GENETICS

A PERIODICAL RECORD OF INVESTIGATIONS
BEARING ON HEREDITY AND VARIATION

Volume 32 JANUARY, 1947 Number 1

TABLE OF CONTENTS

STURTEVANT, A. H., Thomas Hunt Morgan 1

AUERBACH, C., Abnormal segregation after chemical treatment of Drosophila 3

GORDON, MYRON, Genetics of Platypoecilus maculatus. IV. The sex determining mechanism in two wild populations of the Mexican Platyfish 8

KODANI, MASUO, Variations in the terminal bands of the salivary X chromosome of *Drosophila melanogaster* 18

SCHMIEDER, R. G., and P. W. WHITING, Reproductive economy in the Chalcidoid Wasp Melittobia 29

NEEL, JAMES V., and WILLIAM N. VALENTINE, Further studies on the genetics of Thalassemia 38

OWEN, R. D., CLYDE STORMONT, and M. R. IRWIN, An immunogenetic analysis of racial differences in dairy cattle . . 64

SAX, KARL, Temperature effects on X-ray induced chromosome aberrations 75

Abstracts of Papers Presented at the 1946 Meetings of the Genetics Society of America 79

PUBLISHED BIMONTHLY AT MENASHA, WISCONSIN
BY THE BROOKLYN BOTANIC GARDEN
BROOKLYN, NEW YORK, U.S.A.

(Date of issue, January, 20, 1947)

EDITORIAL BOARD

CURT STERN, *Managing Editor*, University of Rochester

WILLIAM E. CASTLE
University of California

LEON J. COLE
University of Wisconsin

EDWIN G. CONKLIN
Princeton University

BRADLEY M. DAVIS
University of Michigan

L. C. DUNN
Columbia University

ROLLINS A. EMERSON
Cornell University

JOHN W. GOWEN
Iowa State College

HERBERT S. JENNINGS
University of California
Los Angeles

DONALD F. JONES
Connecticut Agricultural
Experiment Station

M. M. RHOADES
Columbia University

GEORGE H. SHULL
Princeton University

T. M. SONNEBORN
Indiana University (Representative of the Genetics Society of America)

L. J. STADLER
U. S. Dept. Agriculture
University of Missouri

ALFRED H. STURTEVANT
California Institute of Technology

SEWALL WRIGHT
University of Chicago

GEORGE M. REED, BUSINESS MANAGER, BROOKLYN BOTANIC GARDEN

Volume 32 JANUARY, 1947 Number 1

GENETICS is a bimonthly journal issued in annual volumes of about 600 pages each.

Subscription, $6.00 net a year for complete volumes (January-November). Foreign postage, 50 cents additional. Single copies, $1.25 each, postpaid.

As available, volumes 1 to 6 may be had at $9.00 each, and the later volumes at $7.50 each, postpaid. The Business Manager will supply information on request as to volumes and numbers available.

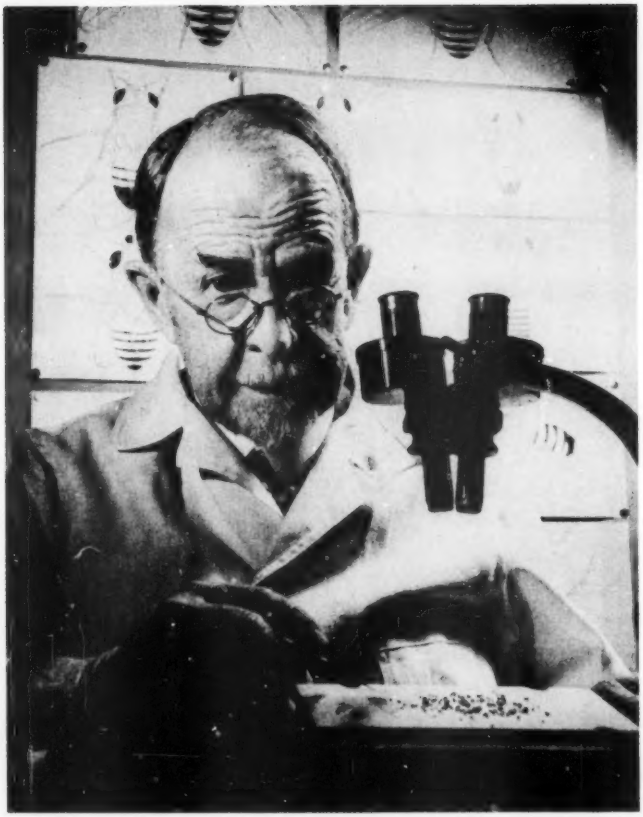
Correspondence concerning editorial matters should be addressed to the EDITOR OF GENETICS, Dewey Bldg., University of Rochester, Rochester, N. Y.

Business Correspondence, including change of address, directions concerning reprints, and exchanges, should be addressed to GENETICS, Menasha, Wisconsin, U.S.A., or BROOKLYN BOTANIC GARDEN, 1000 Washington Ave., Brooklyn 25, N.Y., U.S.A.

Remittances should be made payable to GENETICS.

Entered as second-class matter, August 31, 1922, at the post-office at Menasha, Wisconsin, under Act of March 3, 1879. Acceptance for mailing at the special rate of postage provided for in the Act of February 28, 1925, paragraph 4, section 412, P.L. & R., authorized January 9, 1932.

Claims for missing numbers should be made within 30 days following their date of mailing. The publishers will supply missing numbers free only when they have been lost in the mails.



Thomas Hunt Morgan

THOMAS HUNT MORGAN

THOMAS HUNT MORGAN was born Sept. 25, 1866, at Lexington, Kentucky, the son of CHARLTON HUNT MORGAN and ELLEN KEY (HOWARD) MORGAN. His father was a brother of JOHN HUNT MORGAN of Civil War fame; his mother was a granddaughter of FRANCIS SCOTT KEY (author of "The Star Spangled Banner"), and traced to several of the old aristocratic families of Maryland and Pennsylvania. This background, combined with his sympathetic understanding, lively sense of humor, and complete lack of snobbishness, made MORGAN at ease and a delightful and stimulating person in any company.

MORGAN'S early work, like that of several other early animal geneticists, was in embryology. He was one of the leaders in the development of experimental embryology in the 1890's, and also took the lead in the development of the related subject of regeneration. He continued to work in these fields, and was carrying on experiments in regeneration at the time of his death (Dec. 4, 1945).

MORGAN'S interest in genetics arose primarily from a feeling that the study of mutation offered a way out of the speculative and rather futile trend of the time in the discussion of evolution. As late as 1908 he was still quite critical of parts of the Mendelian interpretation of heredity; but two years later, when his first results with Drosophila mutants were published, these doubts quickly disappeared. He was at this time also engaged in a cytological study of sex determination in the complex life-cycle of the Phylloxerans—a study which furnished perhaps the most brilliant and convincing of the early confirmations of the rôle of the chromosomes in the determination of sex.

Because of this work, and also because he was then (as for so many years thereafter) closely associated at Columbia University with E. B. WILSON, it was natural that MORGAN should turn to the chromosomes for an interpretation of the early results with Drosophila. The discovery of sex-linkage of the type with heterozygous male came in 1910, and was quickly followed by the proof that recombination occurs between sex-linked genes, and that such recombination is not random—i.e., that sex-linked genes are linked to each other. Already in 1911 these results were correlated with chromosome behavior, the phenomenon of crossing over was interpreted in terms of JANSSENS' chiasma-type hypothesis, and it was suggested that closeness of linkage is dependent on nearness together in the chromosomes of the genes concerned. Here, in outline, was the basis of modern genetics.

From this point on in the development of the Drosophila work it becomes difficult to say who was responsible for which advance. MORGAN early developed a group of students, with whom he worked in close contact. BRIDGES, MULLER, and the writer formed with him, in the years immediately following, a closely cooperating group. Discussion was free, each knew what the others were doing, and new discoveries and ideas were frequent. We were bent on getting ahead with the problem, and results were what mattered, rather than

questions of credit or priority—the latter being things in which MORGAN had little interest.

The Drosophila work entered a new phase in 1915, with the publication of "The Mechanism of Mendelian Heredity," the first consistent attempt to present an account of the whole field in terms of the chromosome interpretation. In that same year MORGAN obtained a grant from the CARNEGIE INSTITUTION, which he used to support BRIDGES and the writer as full-time investigators. Our position was unusual, since we were given complete freedom in our work. We called MORGAN "The Boss"—but that was a term of affection and respect, and was not at all intended to suggest that he directed the course of our work or in any way treated us as his assistants.

From this time on MORGAN'S own experimental work with Drosophila gradually became less in amount. He vigorously supported the work of others and played a large part in stimulating it and in the development and presentation of the general conclusions. He was, however, happiest and most efficient when working in less thoroughly exploited fields. He always had numerous experiments under way, involving various kinds of material. Many of these experiments did not yield significant results and were never published; but to this period belong his important work on hen-feathering in fowls, on the determination of the large claw in the fiddler-crab, much of his work on self-sterility in Ciona, and a series of studies on experimental embryology.

MORGAN'S academic career may be summarized as follows: B. S., University of Kentucky, 1886; Ph.D., Johns Hopkins University, 1890; Professor of Biology, Bryn Mawr College, 1891–1904; Professor of Experimental Zoology, Columbia University, 1904–1928; Professor of Biology, California Institute of Technology, 1928–1945. He was President of the National Academy of Sciences (1927–1931), of the American Association for the Advancement of Science (1930), and of the International Genetics Congress (1932). In 1933 he was awarded the NOBEL PRIZE in Medicine. He was a member of the editorial board of GENETICS from its founding until his death.

In 1904 he was married to LILIAN V. SAMPSON, who survives him. Mrs. MORGAN is herself a well-known geneticist, having contributed much to the development of the Drosophila work.

A. H. STURTEVANT

ABNORMAL SEGREGATION AFTER CHEMICAL TREATMENT OF DROSOPHILA

C. AUERBACH

Institute of Animal Genetics, University of Edinburgh, Edinburgh, Scotland

Received June 20, 1946

AMONG the mustard gas compounds which have been shown to possess a high mutagenic capacity combined with the ability to produce chromosome breaks, N-methyl di-(2-chloroethyl) amine, $CH_3 \cdot N(CH_2CH_2Cl)_2$, is one of the most efficient (AUERBACH and ROBSON, in press). In an experiment carried out in 1943, in which wild type males of *Drosophila melanogaster* were exposed to vapors of this substance, there occurred an abnormal and inherited type of segregation which points to an effect of the chemical on the centromere of the treated X chromosome.

The treated males were mated to *ClB/scar* females.[1] One of the Bar daughters from this mating, when mated to a *scar* brother, produced a highly unusual type of progeny. If *t* is used to indicate the treated X chromosome, the data may be summarized as follows:

P_1 *ClB/scar* ♀ ♀ × *t* ♂ ♂
F_1 *ClB/t* ♀ × *scar* brother

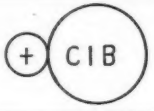
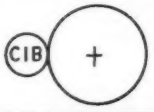
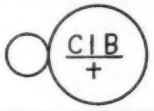
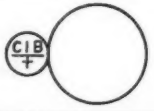
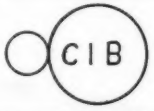
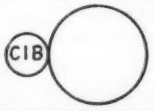
F_2 phenotypes	♀ ♀	♂ ♂
	36 *sc v B*	34 *scar*
	11 wild	3 wild
	9 *B*	1 *sc*
	1 *sc B*	
	57	38

All wild-type males, the *sc* male, and eight tested *scar* males proved to be sterile. The other *scar* males were not tested, but it may be presumed that they, too, were sterile. Sterility of the wild type males may have been caused by a mutation on the treated X chromosome. The same applies to the *sc* male which must have been a crossover between *ClB* and *t*. Sterility of the *scar* males, on the other hand, points to their having lacked a Y chromosome, and thus having been produced by primary nondisjunction in the *ClB/t* female.

Such a high degree of primary nondisjunction is quite unusual. From a study of the segregation data there emerge several points which allow inferences on meiosis in the *ClB/t* female. As is usual with primary nondisjunction, there are

[1] *scar* = *sc*(scute), *v*(vermilion), *f*(forked), *car*(carnation). It may be recalled that the *ClB* chromosome contains the mutant genes *sc*(scute), l^2(tan^2), *v*(vermilion), *sl*(small wing) and *B*(Bar eye).

considerably more exceptional XO males (*scar*) than exceptional XXY females (Bar). This is attributable to the fact that nullo-X eggs, from which the non-disjunctional males arise, may be produced not only by nondisjunction carried to the final result of two X chromosomes becoming included into the same cell,

Type of segregation		Ovule and polar body	Zygote after fertilization by sperm	
			scar	X Y
A	1		*sc v B* ♀ 11 (+|*sc B*)	—
	2		wild ♀ 11	wild ♂ 3 (+|*sc*)
B	1		—	*B* ♀ 9
	2		*scar* ♂ 9	—
C	1		*sc v B* ♀ 25	—
	2		*scar* ♂ 25	—

A-normal segregation. B-non disjunction. C-loss of treated X.

but also by loss of one X chromosome on the spindle. The observed data allow an estimate to be formed of the relative frequencies with which the treated X chromosome (A) segregates normally from the *ClB*, (B) fails to disjoin from the *ClB* and is included with it into the same cell, ovum or polar body, and (C) is lost on the spindle. Figure 1 shows these possibilities in a diagrammatic way.

There are nine Bar females resulting from the inclusion of both X chromosomes into the ovum (B, 1). It may be presumed that there is a corresponding number of nullo-X eggs produced through the inclusion of both X chromosomes in the polar body (B, 2). This, then, accounts for nine out of the 34 exceptional males. The remaining 25 must be due to a combination of segregation of the *ClB* into the polar body and loss of *t* on the spindle (C, 2). This, however, represents only half of the cases of loss of *t;* where this loss is combined with segregation of *ClB* into the ovum, the resulting fly will be a *sc v B* female, (C, 1). Thus there are about 50 cases in which the treated X is lost on the spindle as against about 18 in which it follows the untreated one into the same cell: loss of the treated X is about 2½ times as frequent as nondisjunction.

When the 25 *sc v B* females which may be presumed to have arisen through loss of *t*(C, 1) are subtracted from the total of 36, there remain 11 which arose through normal segregation, corresponding to the 11 wild-type (*scar/t*) females. (A, 1, 2). The shortage of wild type females, like their sterility, possibly was due to mutation on the treated X chromosome. Comparison of A and B shows that normal segregation was about equally as frequent as nondisjunction. This accounts for the two most striking features of the segregation data: the similar frequencies of nondisjunctional males and *sc v B* females, and of nondisjunctional females and wild type females.

Five of the nondisjunctional Bar females of the genotype *ClB/t/Y* were crossed to males *sc v delta 49 od car*.[2] All of them produced mainly nondisjunctional sons and *sc v B* daughters and very little else. The combined data are as follows:

ClB/t/Y ♀ ♀ × *sc v delta 49 od car* ♂ ♂

daughters	sons
77 *sc v B*	80 *sc v od car*
7 wild	3 wild
6 *B*	
90	83

This segregation shows the same features as the one observed in F_2, namely a degree of nondisjunction which is unusual even for secondary nondisjunction in the presence of an inversion, and numerical similarity of patroclinous males with *sc v B* females, and of nondisjunctional B females with wild type females. The only difference from the F_2 is a shift of the frequencies in favor of exceptional progeny, and this is what would be expected from the influence of the extra Y chromosome on meiosis. A similar calculation as has been carried out above gives the following frequencies for the three initial types of segregation: (A) normal segregation: 12, (B) nondisjunction: 14, (C) loss of *t* on the spindle: 174. It will be seen that there has been no relative increase in the frequency with which the treated X chromosome fails to disjoin from the untreated one, but that the relative frequency of its loss has been increased about five times.

[2] delta 49 = large inversion, *od* (outstretched).

Data obtained with breeding from *scar/t* females were of the same type, but too small for numerical interpretation. One out of six mated females differed from all others in producing a progeny with a normal proportion of wild type sons and containing no nondisjunctional individuals. Apparently this female had lost both the semi-lethal and the condition responsible for nondisjunction.

Crossing over took place in all regions separated by the four marker genes of the *sc v f car* chromosome; this shows that no gross rearrangement was present. This was confirmed by an examination of salivary slides kindly carried out by DR. SLIZYNSKI. There appeared to be an abnormality in the region of the centromere, but its nature could not be determined on the basis of the available slides.

Owing to its peculiar behavior the treated X chromosome was rapidly lost: through inviability and sterility of the males carrying it, through loss on the spindle, and through crossing over.

INTERPRETATION

The treated X chromosome was characterized by four peculiarities: it carried a semilethal, it carried a sterility mutation, it had a tendency to follow the untreated X chromosome into the same cell at meiosis, and it had a still greater tendency to be lost at meiosis. The viability and sterility mutation possibly were identical, moreover they must have been located in the same region which carried the cause of the abnormal segregation, for all four peculiarities were lost simultaneously from one female. This region appears to have been the centromere region. It may be considered that under the influence of the chemical treatment a change took place in this region—either a small rearrangement involving the centromere, or a "mutation" of the centromere itself—as a consequence of which the treated chromosome tended to follow its homologue at meiosis on the spindle, either the whole way with inclusion of both in the same nucleus, or part of the way with subsequent loss on the spindle. The presence of an extra Y chromosome does not seem to have influenced the relative frequency of nondisjunction as compared with normal segregation; but presumably through competition for pairing, it appears to have created additional opportunities for loss of the treated X.

Crossing over between the treated X and the *ClB* chromosome seems unusually high: when the nondisjunctional sons, which obviously cannot have received a crossover chromosome, are disregarded, there remain 150 flies in F_2 and F_3 together, and two of these have crossover chromosomes (one single, the *sc* male, and one double, the *sc B* female). The possibility may be considered that crossing over was facilitated through weakening of the centromere.

If the treated X chromosome were lost also during the mitotic divisions, this should lead to a frequent occurrence of male spots in females heterozygous for it. Actually, only one gynandromorph was found among over 50 females which were heterozygous for the treated chromosome. Apparently disturbances of mitosis, if they occurred at all, were infrequent. This suggests that the erratic behavior at meiosis was caused by the unequal strength of the treated centromere and the homologous one. In mitosis, the two centromeres which separate

from one another are the daughters of the same original centromere and must be supposed to be equal in strength independently of whether the mother centromere was normal or abnormal.

Several other cases of abnormal segregation were observed after chemical treatment, but none of them was sufficiently analyzed to suggest the underlying cause. The interest of the present case lies in the fact that it points to the possibility of chemical effects on the centromere. This is in keeping with unpublished results of DR. KOLLER who found definite effects of mustard gas on the centromeres of *Tradescantia* chromosomes.

ANDERSON (1929) has described a line of exceptionally high primary and secondary nondisjunction in *Drosophila melanogaster*. However, the percentages of nondisjunction recorded by him are very much lower than the present ones, and there was no loss of the involved X chromosome on the spindle. This indicates already that the centromere was not responsible, and as a matter-of-fact, ANDERSON was able to locate the cause for the abnormality in the neighborhood of the *v* locus, that is well away from the centromere. Thus, his case seems hardly comparable with the one described here.

LITERATURE CITED

ANDERSON, E. G., 1929 Studies on a case of high non-disjunction in *Drosophila melanogaster*. *Z. i. A. V.*, 51: 397–441.

AUERBACH, C., and J. M. ROBSON (in press). Tests of chemical substances for mutagenic action. Proc. Roy. Soc. Edin.

GENETICS OF PLATYPOECILUS MACULATUS. IV. THE SEX DETERMINING MECHANISM IN TWO WILD POPULATIONS OF THE MEXICAN PLATYFISH[1]

MYRON GORDON

New York Aquarium, New York Zoological Society, New York, N. Y.

Received June 25, 1946

THE genetic mechanism for sex determination in *Platypoecilus maculatus* where ZZ = male and WZ = female was established by BELLAMY in 1922 and confirmed independently by GORDON in 1927. This was more firmly established in 1929 when FRASER and GORDON indicated that crossing over between W and Z occurred. One result of such crossing over was the development of a genetic strain which was characterized by "mother-to-daughter" inheritance; this was effected by W-borne dominant genes *R* and *Sp*, expressed in red body coloring and black spotting. This type of sex determination for the platyfish was confirmed repeatedly by the above workers (BELLAMY 1928; GORDON 1937) and by others, notably KOSSWIG (1938) and BREIDER (1937, 1942).

Recently GORDON (1944) announced that "wild" stocks of *Platypoecilus maculatus* from the Rio Jamapa, Veracruz, Mexico had a type of genetic mechanism for sex determination in which XY = male and XX = female—just opposite to the established type for the species. He also pointed out that the "domesticated" types of the platyfish retained their special type of sex determination in which ZZ = male and WZ = female. When a "wild" female (XX) was crossed with a "domesticated" male (ZZ) all the offspring were males (ZX).

In view of these unusual results a revaluation is necessary of sex determining mechanisms suggested for fishes of the group, the Xiphophorini. In line with the need of clarifying the situation, and substantiating the fact of dual genetic systems in the same species, data will be presented on sex linkage in two "wild" populations, one from the Rio Jamapa and the other from the Rio Papaloapan, Mexico.

MATERIAL

In 1932 one hundred and one *P. maculatus* (platyfish) were caught in a pool of an arroyo leading to the Rio Tonto, a tributary of the Rio Papaloapan in Oaxaca (GORDON, 1940). Within the same pool a smaller number of *Xiphophorus hellerii* (swordtails) were taken. No hybrids between the swordtail and the platyfish have yet been discovered in nature. The fishes were shipped to the genetical laboratory then at CORNELL UNIVERSITY in Ithaca, New York. The following types of platyfish were represented: the spotted-belly (or black-

[1] Aided in part by a grant from the ANNA FULLER FUND and by the facilities of the AMERICAN MUSEUM OF NATURAL HISTORY. The field work in 1939 in Mexico was made possible by a JOHN SIMON GUGGENHEIM MEMORIAL FOUNDATION fellowship.

bottom) controlled by the sex-linked gene *Sb*, the spot-sided, controlled by the sex-linked gene *Sp*, the spotted-dorsal (or black-topper) controlled by another sex-linked gene *Sd*, and the unmarked recessive +. The genetic analysis of these color varieties is given in Table 1.

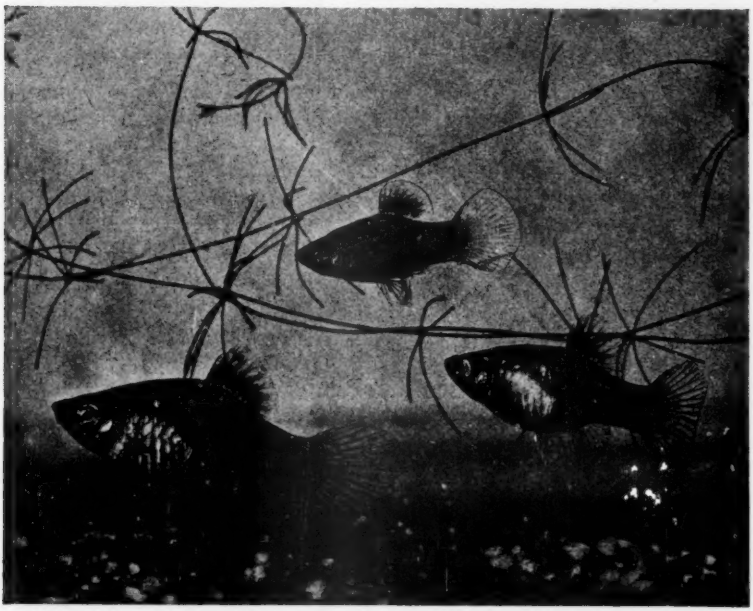


FIGURE 1.—Wild *Platypoecilus maculatus*, the Mexican Platyfish, from the Rio Jamapa, Veracruz, Mexico. This composite photograph shows, on the upper center, a male platyfish having the following sex-linked characters: dorsal-spotted (*Sd*) and stripe-sided (*Sr*); the latter is distinguished by a faint row of macromelanophores along the sides. In the lower left, a female platyfish shows the character spot-sided (*Sp*) prominently and the character stripe-sided (*Sr*) faintly. The female on the lower right is the multiple recessive (+). The other patterns they show, all on, or just in front of, the tail fin are autosomal. The male has the comet (P^{Co}); the left female has the completed crescent (P^{Cc}) and the right female has the comet (P^{Co}) and the one-spot (P^{O}). The sex-linked characters are discussed in this paper; the autosomal, multiple allelic series are discussed in another paper, GORDON (1947). The fish are life size. Photographs by S. C. DUNTON, Staff Photographer, NEW YORK ZOOLOGICAL SOCIETY.

In 1939 on another expedition to Mexico, several thousand platyfish and swordtails were found living side by side in branches of the Rio Papaloapan; no hybrids were found among them (GORDON 1940). At Plaza de Agua, El Tejar, a few miles from Veracruz, a large colony of *P. maculatus* (but no swordtails) was caught and shipped to our laboratory at the New York Aquarium. These represented the platyfish population of the Rio Jamapa system and contained the following sex-linked genes: *Sp*, *Sd*, and *Sr* (striped-sided). All

the sex-linked genes mentioned produce their pattern effects by differential distribution of *macromelanophores*. An analysis of the patterns representative of the Rio Jamapa population is given in Table 2. In addition to the color types mentioned, the platyfish of the two river system have a series of patterns controlled by autosomal genes; an analysis of their genetic behavior is soon to be published (GORDON 1947).

GENETIC ANALYSIS OF THE RIO PAPALOAPAN POPULATION

Only a limited number of tests were made of the Rio Papaloapan population of the platyfish. It was for this reason that results from these tests were not

TABLE 1

Platypoecilus maculatus population from the Rio Papaloapan system.

EXP. NO.	PARENTS				FIRST GENERATION										TOTAL	χ^2
	CULTURE		TYPE		+		*Sb*		*Sp*		*Sd*					
	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂				
1	12	21	+/+	+/*Sb*	20	0	0	15							35	0.706
2	17	21	+/+	+/*Sb*	19	0	0	17							36	0.111
3	2	?	+/*Sp*	(+/*Sb*)	6	0	0	13*	10	0					29	2.310
4	9	?	+/*Sp*	(+/+)	5	4			9	5					23	2.565
5	18	?	+/*Sd*	(+/+)	9	7					5	3			24	3.332

* The 13 fish listed as *Sb* consisted of *Sb* and *Sb*/*Sp* but at the time they were not distinguishable.

given the consideration they deserved at the time they were obtained. In the light of the data obtained later with wild fish from the Rio Jamapa, listed in Table 2, the genetic behavior demonstrated may be explained on the basis of the sex determining mechanism where XX = female, XY = male, and where the Y may carry dominant genes.

In experiment number 1 where the female parent was normally colored, (X)+/(X)+, and the male had the black-bottom pattern, the male presumably carried the gene on its Y chromosome: (X)+/(Y)*Sb* because the dominant *Sb* was transmitted from father to son. This patroclinus mode of inheritance was repeated in the second experiment.

In experiment number 3, a male of the constitution (X)+/(Y)*Sb* must have mated with the spotted female, presumably heterozygous (X)+/(X)*Sp*, to account for the results obtained. Some of the daughters were normally colored (+) while others were spotted (*Sp*). The sons were black-bottomed and presumably about half carried the *Sp* gene in addition to the *Sb*, but at the time the record was made they were all listed as only black-bottomed.

Additional evidence that the *Sp* in females is carried by the X chromosome is given in mating number 4. Here the spotted female presumably was heterozygous (X)+/(X)*Sp*, the male was normally colored (X)+/(Y)+ and the offspring were spotted and non-spotted; in this brood the sexes were equally

distributed among the two phenotypes. Essentially similar results were obtained in the next mating, 5, with the use of the spotted dorsal female (X)+/(X)*Sd*.

Summing up, the Rio Papaloapan fish populations showed that one gene, *Sb*, was transmitted through the Y chromosome and the genes *Sp* and *Sd* were transmitted through the X chromosome. This conclusion, apparently so obvious now, was not appreciated because the bulk of the published data available at the time clearly indicated that WZ = female, ZZ = male.

GENETIC ANALYSIS OF THE RIO JAMAPA POPULATION

A number of young female platyfish were isolated for two or more months. Those which did not produce any offspring within that time were paired with males as indicated in Table 2. In Exp. 1, the spot-sided female transmitted its *Sp* gene (when mated to a recessive male) to half of its daughters and sons; the other half of the offspring, both female and male, were normally colored. These results may be explained on this basis: (X)*Sp*/(X)+ × (X)+(Y)+ = (X)+/(X)+, (X)*Sp*/(X)+, (X)*Sp*/(Y)+, (X)+/(Y)+. These results cannot be explained on the basis of WZ = female, ZZ = male, worked out for this species (*P. maculatus*) from domesticated stocks. If the *Sp* had been borne on the *Z* chromosome, criss-cross inheritance would have been expected; if the *Sp* has been borne on the W chromosome then matroclinus inheritance would have been expected.

The results of Exp. 2 are noteworthy in light of later work, in that they show that a wild platyfish female may carry the *Sd* gene on its X chromosome. One or the other or both of the parents must have been homozygous for the *Sd* gene since all of the offspring showed up with a spotted dorsal fin. At a later point an instance of crossing over of the sex chromosomes (experiment 7) was found in which the *Sd* gene was transferred from the Y to the X.

A first case of a Y borne gene involving *Sd* was detected in mating number 3. Here the male carried *Sr* and *Sd* in this manner: (X)*Sr*/(Y)*Sd*, while the female was homozygous recessive (X)+/(X)+. The female offspring were (X)*Sr*/(X)+ while the males were all (X)+/(Y)*Sd*.

In the 4th and 5th experiments, the male used was homozygous for *Sr*. When mated with an (X)*Sr*/(X)*Sd* female, half of its young were *Sr*, the others were *Sr*/*Sd*; when it was mated with a homozygous recessive female, all its offspring were *Sr*.

The spotted female in mating 6 (1–18) was first mated to an *Sr* male and several broods were produced; then after a two month period of unproductiveness, the same spotted female (X)*Sp*/(X)+ was mated (number 7) to an (X)*Sr*/(Y)*Sd* male. From the second mating (7) its male offspring were (X)+/(Y)*Sd* and (X)*Sr*/(Y)*Sd*; the females were (X)*Sr*/(X)+, (X)*Sr*/(X)*Sp* and an exceptional marked individual, presumably (X)*Sd*/(X)+. This exceptional female (28 *Sd* 1) was tested at a later date (see mating number 14) with a homozygous *Sr* male (30^{3}–11). Since mating number 14 produced sons and daughters in the *Sr*/+ group and in the *Sr*/*Sd* group, the presumed constitution of the exceptional (28 *Sd* 1) female was (X)*Sd*/(X)+; and apparently it

was traceable to a crossover in its male parent (16–12 in mating number 7) in which one (X)*Sr*/(Y)*Sd* germ cell produced an (X)*Sd* gamete, see figure 2.

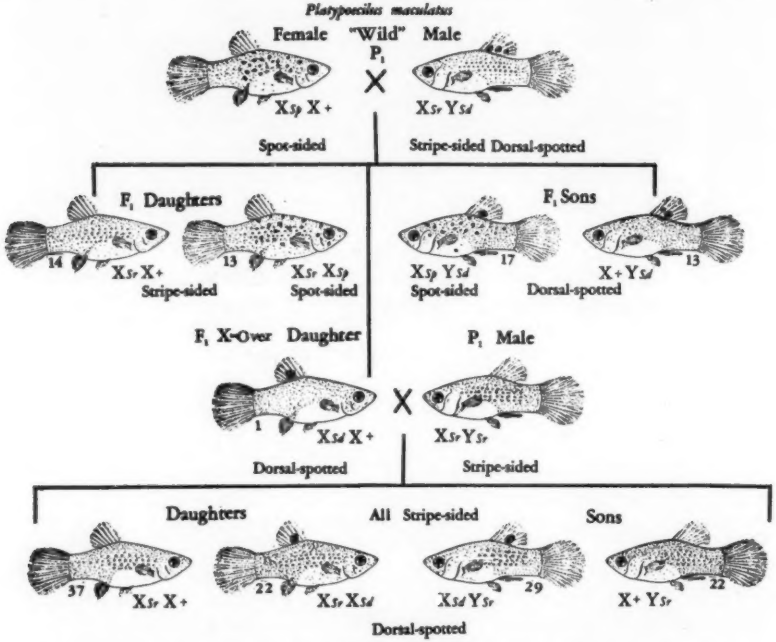


FIGURE 2.—Crossing Over of the X and Y Chromosomes in Mating Number 7. The diagram shows the P_1 male carrying the *Sd* gene on the Y chromosome mated to a *Sp* female. They produced 30 *Sd* sons, 1 *Sd* daughter and 27 non-spotted dorsal daughters. When the exceptional *Sd* daughter was mated to a *Sr* male (Mating Number 14) it produced *Sd* sons and daughters in equal proportions. This supports the supposition that *Sd*, originally on the Y chromosome, was transferred to the X.

The results obtained from matings number 8, 9, 10 and 11 were in accordance with the general results obtained previously where XX = female and XY = male. In these matings no exceptions were detected.

An exceptional individual appeared from mating number 12. Here a female (X)*Sr*/(X)*Sp* was mated to an (X)+/(Y)*Sd* male. Their young were as follows: females = (X)*Sp*/(X)+, (X)*Sr*/(X)+; males = (X)*Sp*/(Y)*Sd*, (X)*Sr*/(Y)*Sd*, and a single exceptional male which was presumably (X)*Sp*/(Y)+. Two explanations may be suggested for its appearance: (1) it may have been the product of crossing over of the sex chromosomes in its male parent (60a14) in which an (X)+/(Y)*Sd* germ cell produced an (Y)+ sperm; (2) it may have represented an incomplete case of sex reversal in which the exceptional male

may not have been of the constitution (X)*Sp*/(Y)+ but (X)*Sp*/(X)+. Since it was sterile, it was not possible to decide just what occurred.

Two more exceptions were found in mating number 13 when an (X)*Sp*/(X)+ female (17–1) was crossed with an (X)*Sr*/(Y)*Sd* male (24–11); they produced two unexpected male offspring presumably having the following constitution: (X)+/(Y)*Sr* which was fertile and (X)*Sp*/(Y)*Sr* which was sterile. The fertile male exception (number 62–12) was mated to a spotted female (159–1) which had the following genetic constitution: (X)*Sp*/(X)+. The results listed in mating number 21 indicate that the *Sr* gene had crossed over from the X to the Y chromosome in male number 24–11 of mating number 13. The results indicate the probable sequence of events as follows:

No. 17-1 (X)*Sp*/(X)+ P_1 × No. 24-11 (X)*Sr*/(Y)*Sd*

F_1

19 (X)+/(X)*Sr*, 24 (X)*Sp*/(X)*Sr*, 20 (X)*Sp*/(Y)*Sd*, 18 (X)+/(Y)*Sd*
1 (X)+/(Y)*Sr**, 1 (X)*Sp*/(Y)*Sr*
Fertile* Sterile

No. 159-1 = (X)*Sp*/(X)+ × (X)+/(Y)*Sr** = No. 62-12

12 (X)+/(X)+, 12 (X)*Sp*/(X)+, 13 (X)+/(Y)*Sr*, 13 (X)*Sp*/(Y)*Sr*

Conclusion: *Sr* crossed over from X to Y in the germ cells of the P_1 male no. 24–11. The exceptional offspring (X)+/(Y)*Sr* (No. 62–12) subsequently transmitted its *Sr* gene to its sons only through the Y chromosome.

Mating number 14 has previously been discussed and numbers 15 and 16 did not produce any unusual results: criss-cross inheritance was observed for *Sp* and *Sr* and patroclinus inheritance for *Sd*. From mating number 17 a remarkable example of sex reversal was found in an exceptional male offspring (GORDON 1946c).

SEXUAL TRANSFORMATION OF A GENETICALLY CONSTITUTED FEMALE INTO A FUNCTIONAL MALE

The events leading to the discovery of a functional male with a genetic constitution of a female are as follows. A stripe-sided female (63–1) was mated to a spot-sided and spotted dorsal male (62–11). This mating and their offspring may be expressed as follows:

(X)*Sr*/(X)+ × (X)*Sp*/(Y)*Sd*

♀♀ F_1 ♂♂

32 (X)*Sr*/(X)*Sp*, 34 (X)*Sp*/(X)+ 29 (X)*Sr*/(Y)*Sd*, 27 (X)+/(Y)*Sd*
1 (X)*Sp*/(X)+* = exceptional ♂

(X)*Sr*/(X)*Sr* × (X)*Sp*/(X)+* = exceptional ♂

F_2

79 ♀♀ (X)*Sr*/(X)*Sp* No males
74 ♀♀ (X)*Sr*/(X)+

TABLE 2

Platypoecilus maculatus population from the Rio Jamapa at El Tejar, Veracruz.

EXP. NO.	PARENTS ♀	PARENTS ♂	X/X	X/Y	PED. NO.	FIRST GENERATION +/+ ♀	+/+ ♂	Sd/+ ♀	Sd/+ ♂	Sp/+ ♀	Sp/+ ♂	Sr/+ ♀	Sr/+ ♂	Sd/Sp ♀	Sd/Sp ♂	Sd/Sr ♀	Sd/Sr ♂	Sp/Sr ♀	Sp/Sr ♂	TOTAL	χ^2
1	1-7	1-17	*Sp*/+	+/+	17	16	13			12	10									51	1.961
2	1-10	1-20	*Sd*/+	*Sd*/*Sd*	20			23	15											38	1.684
3	1-12	1-22	+/+	*Sr*/*Sd*	22			0	14			18	0							32	0.500
4	1-16	1-34	*Sr*/*Sd*	*Sr*/*Sr*	26							13	14			13	8			48	1.833
5	1-17	1-34	+/+	*Sr*/*Sr*	27							25	25							50	0.000
6	1-18	16-11	*Sp*/+	*Sr*/+	28a	0	7			0	5	6	0					8	0	26	0.746
7	1-18	16-12	*Sp*/+	*Sr*/*Sd*	28b			1	13			14	0	0	17			13	0	58	0.719*
8	1-19	1-36	*Sr*/+	+/*Sr*	29	12	0					11	17							40	0.950
9	27-1	27-11	*Sr*/+	+/*Sr*	30	7	0					14	22							43	2.872
10	27-2	12-11	*Sr*/+	+/*Sd*	60a	16	0	0	16			12	0			0	14			58	0.759
11	27-3	21-11	*Sr*/+	+/+	60b	9	9					6	5							29	1.513
12	28a3	60a14	*Sr*/*Sp*	+/*Sd*	61					12	1	16	0	0	11	0	16			56	1.473*
13	17-1	24-11	*Sp*/+	*Sr*/*Sd*	62			0	18			19	1	0	20			24	1	83	1.047*
14	28Sd1	30[2]-11	*Sd*/+	*Sr*/*Sr*	63							37	22			29	22			110	4.654
15	60a3	28b11	+/+	*Sp*/*Sd*	64			0	9	8	0									17	0.059
16	60a5	60a11	+/+	*Sr*/*Sd*	65			0	16			18	0							34	0.118
17	63-1	62-11	*Sr*/+	*Sp*/*Sd*	159			0	27	34	1					0	29	32	0	123	0.950*
18	63-2	63-12	*Sd*/*Sr*	*Sd*/*Sr*	160			18	0			1	12			12	17			70	3.337*
19	61-1	28b17	*Sp*/+	+/*Sd*	161	15	0	0	12	12	0			0	13					52	0.461
20	64-1	63-11	*Sp*/+	*Sd*/*Sr*	163			5	0			0	6	6	0			0	6	23	0.130
21	159-1	62-12	*Sp*/+	+/*Sr*	178	12	0			12	0	0	13					0	13	50	0.082
22	30[4]-1	159-21	*Sr*/*Sr*	*Sp*	167							79	0					74	0	153	0.163

* The values starred (*) were obtained by omitting the exceptional cross-overs.

A single spotted-male appeared in the F_1. This phenotype generally characterized the females. When the exceptional spotted male (159–21) was tested by mating it with a homozygous stripe-sided female (30⁴–1), the pair produced 153 females and no males (see mating 22). A histological report on the reproductive organs of male number 159–21 is being prepared. The female number 30⁴–1 with which 159–21 was mated, after a period of non-productivity, was mated a second time with an (X)+/(Y)+ male. From the latter mating (not indicated in Table 2) it produced approximately as many male as female offspring. This definitely shows that the first male, number 159–21, was responsible for the unusual sex ratio in the first group of young.

In mating number 18 another exceptional individual was discovered; this time it was a female offspring. The simplest explanation for its appearance is that it represented a product of crossing over of the (X)*Sd* and (Y)*Sr* chromosomes of its male parent (63–12): an (X)*Sr* sperm was produced. This suggestion could not be verified owing to the complete sterility of the exceptional female.

Matings number 19 and 20 may be contrasted with regard to the manner in which the gene *Sd* was carried by the male parents. In 19, the male (28b17) *Sd* was carried by the X chromosome whereas in the latter (male, 63–11) *Sd* was carried by the Y. The results are explainable on this basis.

Matings number 21 and 22 were discussed previously in connection with exceptional individuals, 62–12 and 159–21.

DISCUSSION

A general discussion of the genetic sex determining mechanism in "wild" and in "domesticated" stocks of *Platypoecilus maculatus* is presented in another paper by the author (GORDON 1946b). In this paper the consequences of mating a homozygous "domesticated" platyfish male (ZZ) with a homozygous "wild" female (XX) are given: all the offspring are male (ZX). This result and others substantiate the suggestion of CASTLE (1936) that perhaps the usage of ZZ for the "domesticated" male might be discontinued and YY substituted. Apparently Z and Y are homologous. However the W chromosome of the "domesticated" female is not homologous to any X of the platyfish. Indeed, the nature of the W chromosome is not yet clear. GORDON has suggested that perhaps the W chromosome may be traced to a *Xiphophorus hellerii* chromosome which has introgressed through intergeneric hybridization into the genetic structure of the domesticated platyfish. The basis for this suggestion may be found in the results of mating an (X)*Sr*/(Y)*Sd* "wild" male *P. maculatus* with a "wild" female *X. hellerii*. All the *Sr* hybrids are female and fertile; some of the *Sd* hybrids are male, some are female while most of them are sexually indifferent and, of course, sterile. This is as far as the experiment has gone at present. Suppose, however, that the rare fertile male and the rarer fertile female *Sd* platyfish-swordtail hybrids are inbred:

	♀♀	F_1	♂♂
	(X′)+/(Y)*Sd*	×	(X′)+/(Y)*Sd*

F_2:	1 (X′)+/(X′)+	2 (X′)+/(Y)*Sd*	1 (Y)*Sd*/(Y)*Sd*
	presumably female,	presumably female or male,	presumably male

In the second generation hybrids we would probably find some homozygous females containing two X′ chromosomes from *Xiphophorus hellerii*. And we would probably find some homozygous males carrying two Y chromosomes, from *Platypoecilus maculatus*. The others would probably be male or female and most of them might be sterile. If among the latter, an (X′)+/(Y)*Sd* female stock could be stabilized, then the so-called WZ = female and ZZ = male platyfish stock could be reestablished.

Under restricted aquarium conditions *Xiphophorus hellerii* and *Platypoecilus maculatus* hybridized readily and the history of the first reported hybrid between them goes back to 1912. The records show that the first platyfish were imported into Germany in 1907 and the first swordtails in 1909. Under natural conditions, and this phase of research is described by GORDON (1947), the species have never been known to hybridize.

Under commercial tropical fish breeding practices, there are many examples of the transference of platyfish genes into the germ plasm of a swordtail-like hybrid and the subject of introgressive hybridization is discussed by GORDON (1946a).

Both the WZ-ZZ and the XX-XY systems are stable in their respective stocks and are being maintained side by side. According to BREIDER (1942) the stocks maintained by him, at least up to 1942, were still showing the WZ-ZZ mechanism. Indeed, in a remarkable cross he identified an exceptional male platyfish, which by its peculiar phenotype should have been a female. When he tested the unusual male which he suspected had the genetic constitution of (W)*RSp*/(Z)*Dr* with a stock female (W)+/(Z)*R*, he obtained 51 females and 13 males,—that is, 3 females to 1 male. Among the females he claimed to have obtained WW females. This was highly probable but it would have been desirable to have had additional information on the genetical behavior of the WW females. If WW females had been mated with ordinary ZZ males, all of their offspring would have had to be males; this would have verified the genetic constitution of the presumed WW females completely.

The ratio of 3 females to 1 male has been obtained in this laboratory by a method which did not depend upon the sexual transformation of a genetically constituted female into a functional male. A spotted "domesticated" female (W)*Sp*/(Z)+ was mated to a stripe-sided "wild" male, (X)+/(Y)*Sr*. The spotted F_1 hybrids were all females and presumably, (W)*Sp*/(X)+ or (W)*Sp*/(Y)*Sr;* the non-spotted F_1 hybrids were males, half of which were stripe-sided, (Z)+/(Y)*Sr*, and half were normal. (Z)+/(X)+. When some of the spotted F_1 females were tested with "wild" stripe-sided males the following results were obtained:

Mating I			Mating II		
(W)*Sp*/(X)+	×	(X)+/(Y)*Sr*:	(W)*Sp*/(Y)*Sr*	×	(X)+/(Y)*Sr*:
Daughters:		Sons:	Daughters:		Sons:
(W)*Sp*/(X)+		(X)+/(Y)*Sr*	(W)*Sp*/(X)+		(Y)*Sr*/(Y)*Sr*
(W)*Sp*/(Y)*Sr*			(W)*Sp*/(Y)*Sr*		(Y)*Sr*/(X)+
(X)+/(X)+					

When a WY female was mated with an XY male the sex ratio was 1:1 (mating II). When a WX female was mated with an XY male (mating I) the sex ratio was 3 ♀♀ to 1 ♂♂. A complete report on this mating and other "domesticated-wild" matings will appear in the near future.

SUMMARY

The sex determining mechanism in two wild populations of the Mexican platyfish, *Platypoecilus maculatus*, one from the Rio Papaloapan and the other from the Rio Jamapa may be expressed as follows: XX = female, XY = male.

The domesticated stocks of the same species have a sex determining mechanism which may be expressed as follows: WZ = female, ZZ = male. It is suggested, in view of the probable origin of this mechanism from the wild type that the formula might better be written: WY = female, YY = male.

An instance of sexual transformation of a genetically constituted female of the "wild" stock XX has been reported. When this XX male was mated with a normal "wild" type female, also XX, all the offspring, 153 in all, were females, XX.

LITERATURE CITED

BELLAMY, A. W., 1922 Sex-linked inheritance in the teleost, *Platypoecilus maculatus*. (Abstract) Anat. Rec. **24**: 419–420.

1928 Bionomic studies on certain teleosts (Poeciliinae) II. Color pattern inheritance and sex in *Platypoecilus maculatus*. Genetics **13**: 522–530.

BREIDER, HANS, 1937 Gen und Genotypus. Nach Untersuchungen an lebendgebärendern Zahnkarpfen. Der Züchter **60**: 70–77.

1942 ZW-Männchen und WW-Weibchen bei *Platypoecilus maculatus*. Biol. Zbl. **62**: 187–195.

CASTLE, W. E., 1936 A simplified explanation of Bellamy's experiments concerning sex determination in tropical fishes. Proc. Nat. Acad. Sci. **22**: 679–682.

FRASER, A. C., and MYRON GORDON, 1929 The genetics of Platypoecilus. II. The linkage of two sex-linked characters. Genetics **14**: 160–179.

GORDON, MYRON, 1927 The genetics of a viviparous top-minnow Platypoecilus; the inheritance of two kinds of melanophores. Genetics **12**: 253–283.

1937 Genetics of Platypoecilus. III. Inheritance of sex and crossing over of the sex chromosomes in the platyfish. Genetics **22**: 376–392.

1940 The fish that was jungle born and city bred. Natural History **45**: 96–106.

1944 The XY sex-chromosomes determine maleness in wild populations of *Platypoecilus maculatus* while WZ determine femaleness in domesticated breeds of the same species (abstract). Rec. Genet. Soc. Amer. **13**: 18–19.

1946a Introgressive hybridization in domesticated fishes. I. The behavior of comet, a *Platypoecilus maculatus* gene in *Xiphophorus hellerii*. Zoologica, **31**: 77–88.

1946b Interchanging genetic mechanisms for sex determination in fishes under domestication J. Hered. **37**: 307–320.

1946c Sexual transformation of a genetically constituted female fish (*Platypoecilus maculatus*) into a functional male. (Abstract) Anat. Rec. **94**: 6.

1947 Speciation in Fishes. Distribution in time and space of seven dominant multiple alleles in *Platypoecilus maculatus*. Advances in Genetics **1**: 95–132.

KOSSWIG, CURT, 1938 Über einen neuen Farbcharakter des *Platypoecilus maculatus*. Istanbul Universitesi Fen Fakultesi Mecmuasi **3**(4): 1–8.

VARIATIONS IN THE TERMINAL BANDS OF THE SALIVARY X CHROMOSOME OF *DROSOPHILA MELANOGASTER*

MASUO KODANI[1]

Department of Zoology, University of Rochester, Rochester, N. Y.

Received July 7, 1946

IN SEVERAL normal and spontaneous mutant strains of *Drosophila melanogaster* a varying number of bands appear to be absent in the distal half of the 1A section (BRIDGES' reference map, 1938) of the X chromosomes of the salivary gland nucleus. Studies on these chromosome tips in the stocks of these strains as well as in various hybrids of these strains have revealed that some of the "absent" bands appear under certain conditions in both the stocks and hybrids. It is the purpose of this communication to describe this phenomenon in detail and discuss its meaning.

MATERIAL AND METHODS

The strains used for the present studies are: *yw*, *Rci-4F*, *sn*^{3}, Canton-S, *y* and *Rci-4P*. The two *Rci* stocks are X-rayed rearrangement stocks containing translocations of the chromosomes 4 and were originally isogenic with the Canton-S except for chromosome 4. The following observations were made on aceto-orcein smear preparations of salivary glands in the male and female larvae taken at full maturity from the laboratory stocks of these strains.

In the following description a band which is not visible will be indicated by omission of the number corresponding to that band in the reference map of BRIDGES. Thus, 12345 stands for a complete chromosome in the unpaired condition from male larvae, 12345/12345 for paired complete chromosomes from female larvae, 2345 for an unpaired chromosome from male larvae, with a band at the 1A1 position not visible and 2345/345 for paired chromosomes of female larvae in which a band at the 1A1 position is absent in one and the bands at the 1A1 and 2 positions are absent in the other chromosome.

OBSERVATIONS

Homo- and hemizygotes

yw

About 500 X chromosome pairs in ten pairs of glands from female larvae and about an equal number of X chromosomes also in ten pairs of glands from male larvae were studied, and in every chromosome in both cases all the bands in the terminal region were present (fig. 1a).

4F (*Rci-4F*)

All of 276 chromosome pairs (in six pairs of glands from female larvae) were 2345/2345 (fig. 1b) except one, which was 345/345 (fig. 1d). It is evident that

[1] Supported in part by a grant from the ROCKEFELLER FOUNDATION for assistance to PROFESSOR CURT STERN.

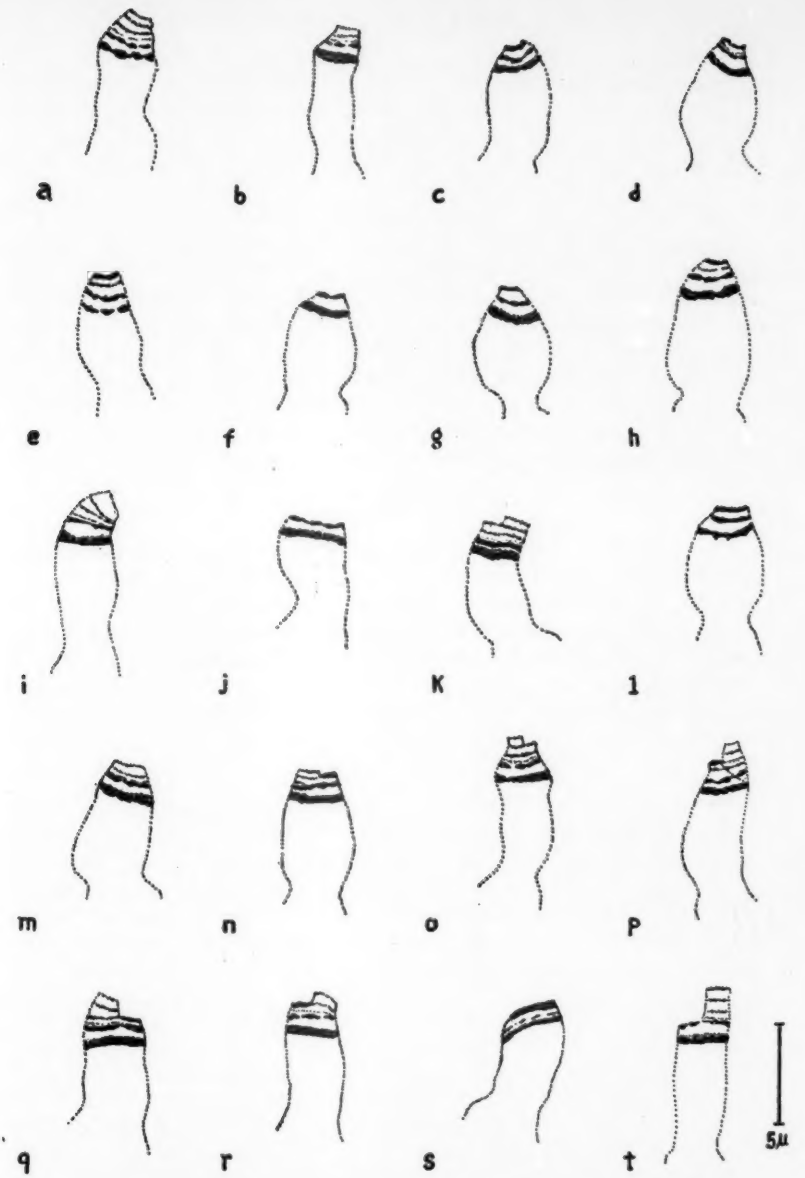


FIGURE 1.—The camera lucida drawings of the tips of the X chromosomes showing variations in configuration in the various strains and hybrids. The lowermost thick band is 1B1234 and the thick band immediately above is 1A56. a—*yw* (unpaired). b—$4F/4F$. c—sn^3/sn^3. d—$4F/4F$. e—sn^3/sn^3. f—y/y. g—y/y. h—y/y. i—y/y. j—$4P$ (unpaired). k—$4F/yw$. l—$4F/+^C$. m—$4F/+^C$. n—$4F/+^C$. o—$yw/+^C$. p—$yw/+^C$. q—yw/y. r—$y/+^C$. s—$y/4P$. t—$yw/4P$. Scale in t is for all figures.

the 1A2 band nearly always appears when the chromosomes are paired. In chromosomes of the male this band appeared very frequently in most of the glands. Of six pairs of glands and 335 chromosomes studied, in five pairs all the chromosomes were 2345, whereas in one pair 31 chromosomes were 2345 and 16 were 345.

sn^3

Among the chromosomes in six pairs of glands, 60 were 2345/2345 (fig. 1e), 45 were 345/345 (fig. 1c), and three were 12345/12345. Most of the X chromosomes of the male (129 in six pairs of glands) were 2345, whereas a small number (ten chromosomes) were 345. The latter condition was found only in one pair of glands. Among the paired X chromosomes of the females the first two types were found in every pair of glands, whereas the 12345/12345 type was found only in one of the six pairs. The 1A2 band in 2345/2345 or 2345 and the 1A3 band in 345/345 are distinctly thicker and more chromatic than usual. The 1A3 band in 2345/2345 or 2345 is very thin and stains faintly.

+^{C} (*Canton-S*)

About 500 X chromosome pairs of the females and 500 X chromosomes of the males, each group from eight pairs of glands, were studied. Almost all the chromosomes were of the same type—that is, 345/345 in the female and 345 in the male chromosomes. One 2345/2345 chromosome pair was found. This exception indicates that the 1A2 band can make its appearance, although it is usually not observed in these chromosomes. The terminal 1A3 band in both 345/345 and 345 is very thick and highly chromatic as is the 1A2 band of the 2345/2345 type.

y

Thirteen pairs of glands taken from female larvae and eight pairs of glands from male larvae were studied. Among the X chromosome pairs of the female were found five types—namely, 12345/12345, 2345/2345, 345/345, 45/45 and 5/5. Among those of the male were found only three types—345, 45, and 5. The actual numbers of these types found in both male and female larvae are indicated in table 1. In the 345/345 type of chromosome the 3-band was generally very thick and highly chromatic and the 4-band, in all the chromosomes, was very thin and feebly stained (fig. 1h). In the 45/45 type of chromosomes a very thick and highly chromatic band was present distal to the 1A5 band without a thin band in between the two (fig. 1g). This terminal band cannot be the 1A4 band because of its unusual thickness and chromaticity. This thick band is interpreted as the compound band consisting of all four bands distal to 1A5. The basis for this interpretation will be discussed later. This type was classified as 45/45 or 45 and counted together with 345/345 and 345. The table reveals the following facts: (1) In the paired chromosomes the proportion of the 345/345 and 45/45 to 5/5 (fig. 1f) varies considerably among the 13 pairs of glands. In most cases the 345/345 and 45/45 types are proportionally larger than the 5/5 type. (2) In the unpaired chromosomes the 345 type appears only

in a small number of larvae, and in those in which it appears at all the frequency is very small. It is clear that the 1A3 band appears with a significantly higher frequency in chromosomes of the female than in chromosomes of the male. (3) The appearance of the 2345/2345 type of chromosomes in three pairs of the female glands and of one chromosome pair of the 12345/12345 type (fig. 11)

TABLE 1

The actual numbers of the various types of the X chromosome tips from male and female larvae in the yellow strain. For details see text.

X CHROMOSOMES FROM 8 MALE LARVAE			
	345 & 45	5	
	0	37	
	0	16	
	0	22	
	2	13	
	2	21	
	0	25	
	1	15	
	0	16	
X CHROMOSOMES FROM 13 FEMALE LARVAE			
12345/12345	2345/2345	345/345 & 45/45	5/5
	2	2	28
		24	12
		6	22
	3	35	12
		25	4
		15	23
		36	18
		24	22
1		21	30
		52	34
		29	7
		21	11
	3	37	10

in one pair of the glands indicates that under some conditions the 1A1 and 2 bands can appear, and the fact that these types were found in the females but not in the males seems to indicate that the pairing is the cause of the appearance of these bands. A number of cases among the paired yellow chromosomes in which the tip ends of the paired chromosomes are unpaired have been observed, and in no case was the 1A3 band present in either one of the unpaired chromosome tips. These cases and similar cases found in other X chromosomes indicate that the higher frequency of the appearance of a certain band such as the 1A3 in the yellow chromosome in paired chromosomes than in unpaired chromosomes is due to the pairing of the 1A section of the chromosomes.

4P (Rci-4P)

All the chromosomes observed in six pairs of glands each of female and of male larvae were 5/5 and 5 (fig. 1j).

The frequencies of the various types of configurations described above are summarized in table 2. The figures in the table represent the frequencies in percentage of the appearance of the five bands in each strain.

TABLE 2

Frequencies with which the four bands at the terminal region of the X chromosome were observed in the various strains are shown in percentage. Note that the frequencies are greater for certain bands in some strains when the chromosomes are paired (female) than when not paired (male) as, for example, in the case of the 1A 3 band in the yellow strain. For further details see text.

	1A5	1A4	1A3	1A2	1A1	TOTAL NO. OF OBSERVED CHROMOSOMES
4P	100					200 ca.
4P/4P	100					200 ca.
y	100	100	3			170
y/y	100	100	58.57	1.57	0.17	569
$+^{c}$	100	100	100			500 ca.
$+^{c}/+^{c}$	100	100	100	0.2 ca.		500 ca.
4F	100	100	100	9.2		335
4F/4F	100	100	100	99.6		276
sn^{3}	100	100	100	92.1		129
sn^{3}/sn^{3}	100	100	100	59	4	108
yw	100	100	100	100	100	500 ca.
yw/yw	100	100	100	100	100	500 ca.

A similar study was made for various hybrids of the strains used in the above study, and the following results were obtained from F_1 hybrid females:

4F/yw

The majority of the chromosomes were 12345/2345 (fig. 1k). The 12345 chromosome is probably the *yw* chromosome and the 2345 chromosome, the *4F* chromosome, since in the *yw* or *yw/yw* chromosome all five bands are always visible, whereas in *4F* or *4F/4F* chromosome 1A1 is constantly absent. A very small number of chromosomes were found to be 12345/345. The latter chromosome of the pair is probably the *4F* chromosome for the same reason. Comparison of the frequencies of the appearance of 1A2 in *yw/4F* and *4F/4F* indicates no significant difference between the two.

4F/+^{c}

Six pairs of glands were studied and in three of these, more than 90 percent of the observed chromosomes were 2345/2345 (fig. 1m), whereas the rest of the observed chromosomes were 2345/345 (fig. 1n). In the other three pairs of

glands 60 percent or more of the observed chromosomes were 2345/345, whereas most of the remaining chromosomes were 2345/2345 and a very small number was 345/345 (fig. 1l). The appearance of the 1A2 band is clearly very much more frequent in the $+^{c}$ chromosome when it is paired with the *4F* chromosome than with another $+^{c}$ chromosome.

4F/y

In every one of seven pairs of glands the majority of the observed chromosomes were 2345/345, and a very small number of chromosomes were 2345/5 or 345/5. As indicated in table 1, the frequency of 345/345 in *y/y* varies considerably among the glands, being very small in some glands. In *4F/y*, on the other hand, the 2345/345 type was found to be uniformly very frequent, indicating that the 1A3 band appears in the yellow chromosome more frequently when this chromosome is paired with *4F* chromosome than when it is paired with another yellow chromosome. (In this type the 2345 and 345 chromosomes are probably the *4F* and yellow chromosome, respectively, since 1A2 is present far more frequently in the *4F* chromosome than in the *y* chromosome.)

4F/4P

All the observed chromosomes in four pairs of glands were 2345/5, where the shorter chromosome is probably the *4P* chromosome, since *4P* is always of the 5 type whereas *4F* is always longer than this type.

$+^{c}$/*yw*

The majority of the observed chromosome pairs in seven pairs of glands was 12345/345 (fig. 1p) and a very small number of the chromosomes was 12345/2345 (fig. 1o). In both cases the longer chromosome is likely to be the *yw* chromosome, since the *yw* chromosome was found previously to be constantly 12345, whereas the $+^{c}$ chromosome was never of this type.

$+^{c}/y$

In four pairs of glands studied, two types, 2345/345 and 345/5, were found. The ratio of these two types in each pair of glands were 30:25, 60:0, 40:10 and 45:12. The 2345/345 type (fig. 1r) is more frequent than the 345/5 type. Which one of the pair in each of these types is the $+^{c}$ chromosome cannot be ascertained However, in the 345/5 type the 345 chromosome is likely to be the $+^{c}$ chromosome, since the $+^{c}$ chromosome was found previously to be almost always 345, whereas the yellow chromosome was found to be often 5.

y/yw

Two hundred chromosomes in four pairs of glands were observed. The majority of these chromosomes were clearly 12345/5. In a small number of chromosomes the configuration was 12345/345 (fig. 1q). In both types of configuration the shorter chromosome is considered to be the *y* chromosome, because 1A1 has never been observed in *y* or *y/y*.

y/4P

All the observed chromosomes in four pairs of glands were 5/5 (fig. 1s).

yw/4P

All chromosomes are 12345/5 (fig. 1t) where the shorter chromosome is considered to be the *4P* chromosome, because in *4P* and *4P/4P* chromosomes the configuration was always the 5 and 5/5 types, respectively.

CONCLUSION

To summarize, the following general statement may be made: (1) Varying numbers of terminal bands appear to be absent in the X chromosome of *4F*, *sn*^{3}, $+^C$, and yellow stocks. In the *4P* stock the 1A1, 2, 3, and 4 bands have not been observed. (2) In the X chromosome of the yellow stock the apparently absent bands appear more frequently when it is paired than when it is not paired, indicating that the pairing may increase the length of the salivary X chromosome. (3) This pairing effect is pronounced in some hybrids—namely, *4F*/$+^C$ and *4F*/yellow. Thus the 1A2 band appears in $+^C$ chromosome more frequently in *4F*/$+^C$ than in $+^C/+^C$, and the 1A3 band appears in the yellow chromosome more frequently in *4F* than in *y/y*.

DISCUSSION

In the *sn*^{3} chromosome, in both paired and unpaired conditions, the 1A3 band is very thick and highly chromatic when this is the terminal band. The 1A2 band is likewise very thick and intensely stainable when this band is at the terminal position. In this case the 1A3 band is thin and only feebly stainable. When all three bands, 1, 2, and 3, are present, these bands are all thin and faintly stainable. A simple interpretation of these cases is that the 1A3 band at the terminal position is a compound of the 1, 2, and 3 bands and the 1A2 band at the terminal position consists of two bands, 1 and 2. The compound nature would explain the greater thickness and higher chromaticity of these bands as well as the fact that these bands are reduced in thickness and chromaticity when another band appears distal to them, since this distal band is considered to be separated from the compound band, leaving in its position a single band. A similar explanation can be given to other cases such as the *4F*, $+^C$ and *y* chromosomes. We may also deal here with the case of the 1A4 band previously mentioned. It was pointed out that a thin and faintly chromatic band appears at the 1A4 position only when one or more bands are present distal to the 1A4 position and that in some cases a thick and highly chromatic band is present in the 1A4 position. This thick band at the 1A4 position would be a compound band consisting of the bands 1, 2, 3, and 4, since all these bands were observed in this chromosome. If it is considered that the bands 1, 2, and 3 separate as a group from the band 4, the results would be a thick compound band at the 1A3 position and a single thin band at the 1A4 position which would be the true 1A4 band. The compound 1A3 band may separate into a compound 1A2 band consisting of the bands 1 and 2 and the single 1A3 band. What would correspond to this type was actually found in

some paired *y* chromosomes (2345/2345 in table 1). The compound 1A2 band may divide into single 1A1 and 1A2 bands (12345/12345).

The process explained above is shown diagramatically in figure 2. This hypothetical scheme is based on the assumption that at the earliest stage of the salivary chromosome development the four bands at the terminal region of the

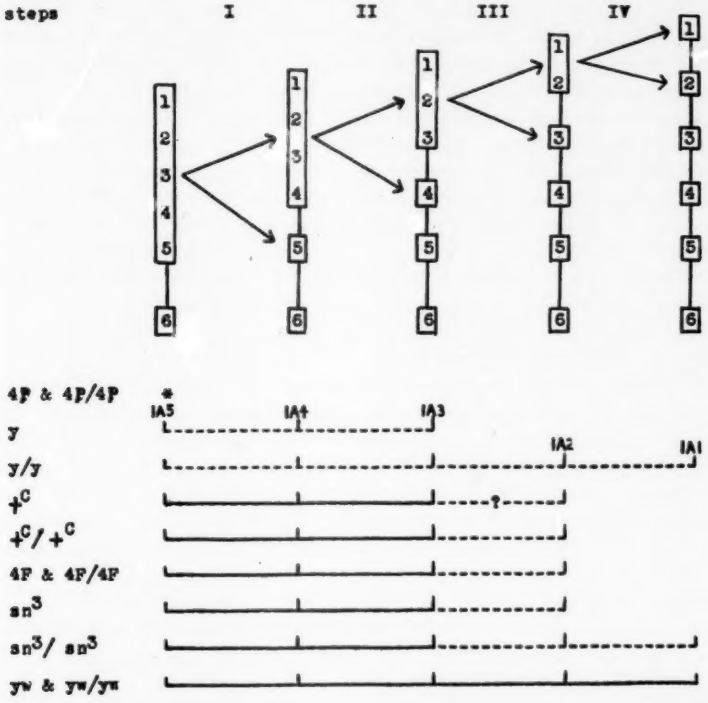


FIGURE 2.—The process of elongation of the terminal region of the X chromosome suggested in the text is diagramatically explained in the upper half of the figure. A block with one number indicates one band corresponding to that number in the reference map of BRIDGES. A block with more than one number indicates a compound band consisting of the bands numbered in the block. Arrows indicate separations of the compound bands into constituent bands. At the lower half of the figure are shown the extent of the separation of the bands in each strain. For details see text.

X chromosome are included in a compound chromomere in the early prophase type of chromosome from which the definitive salivary chromosome develops and that during its development these four bands are segregated out from this compound chromomere. The complete separation of the bands requires four steps which are indicated above the chromosome diagrams. The steps through which the tips of the X chromosome of the various strains pass are indicated

by the horizontal lines at the lower part of the figure. The solid lines indicate the steps through which the tips of these strains pass before the larvae reach their full maturity, and the broken lines represent the extent of variation in the steps through which these tips may pass in addition to those indicated by the solid lines. Studies of the chromosomes in younger larvae might indicate what actually happens, but the difficulty of obtaining adequate smear preparations from very young larvae hinders a study of this small localized area in detail in the earlier stages of development. Actual studies of early third instar larvae of the *y* stock show that the condition of the X chromosome tips is essentially like that in the fully grown larvae.

It may be assumed, on the other hand, that the four bands at the terminal region of the X chromosome constitute separate chromomeres at a very early stage of development of the salivary chromosome. Under this assumption, the compound bands described above would be derived by coalescence of these chromomeres. This is the reverse of the process which is generally believed to be the cause (at least partly) of an enormous elongation of the salivary chromosome during its development from a relatively short prophase type of chromosome. Since there is no reason to assume that the reverse process occurs at the terminal region of the chromosome, it is considered as a less probable process than the one shown in figure 2.

There have been found a few cases in which the band next to the terminal band is more chromatic than the latter. For example, a case was found in the paired sn^3 chromosomes of 2345/2345 type, where the terminal 1A2 band was thin and faintly stained, whereas the 1A3 band was thick and intensely stained. According to the process illustrated in figure 2, this would mean that when these chromosomes underwent the step III, the 1A1 band was separated from the compound band consisting of 1A1, 2, and 3, forming a thin terminal band, while the 1A2 and 3 bands remained together as a thick and intensely stainable compound band at a subterminal position. Cases of this type have been found only very rarely. Incidentally, the case of this type definitely refutes a possibility that the greater chromaticity of the terminal bands is due not to the compound structure but rather to the terminal position which may provide these bands with more staining surface than that of the interstitial bands. The mechanism underlying the step by step separation of the compound bands illustrated in figure 2 is not clear. One, and probably the simplest, possibility is that the elongation starts, in the interband connecting threads at the most proximal position—that is, in the thread between the bands 1A4 and 5—and progresses distally through successive interband threads.

The fact indicated in table 1, that the compound 1A3 appears more frequently in the paired *y* chromosomes, would mean, according to the step by step separation of the compound bands, that the additional steps I and II occur more frequently in the paired condition than in the unpaired condition of these chromosomes. Furthermore, the fact that the 1A2 band appears more frequently in the $+^c$ chromosomes when these chromosomes are paired with the 4^F chromosome than when these are paired with $+^c$ chromosomes would mean that the additional step III occurs more frequently in the $+^c$ chromo-

somes in the $+^{c}/4F$ combination than in the $+^{c}/+^{c}$ combination. Similarly, steps I and II occur more frequently in the *y* chromosomes when these chromosomes are paired with the *4F* chromosomes than with the *y* chromosomes. If the mechanism underlying the process were the successive progression of the elongation in the interband connecting threads, the above facts could be interpreted as indicating that the rate of progression varies from strain to strain and that the rate in one strain may be increased to different degrees by different strains. For example, the *4F* chromosome increases the rate in $+^{c}$ and *y* chromosomes more than $+^{c}$ does to $+^{c}$ or *y* does to another *y*.

The results of the present study indicate that for the salivary analysis of the terminal region of the X chromosome, caution is required in selecting a standard X chromosome and in interpreting the absence of bands that may be observed. For a standard chromosome the *yw* type of chromosome must be used (the much used *Indl-49 y Hw* m^2 g^2 chromosome is of the *yw*-type). SUTTON (1940) found an X chromosome of an irradiated Swedish-b fly, which was cytologically deficient for the 1A1 and 2 bands and phenotypically yellow (stock *Df(1) 260-10, y*) and also an X chromosome in the $\widehat{yy}/gB$ stock which was cytologically deficient for the same two bands but phenotypically not yellow (stock *Df(1)260-19*). SUTTON considered that in the first case a single hit broke the two terminal bands off at the same time it produced a point mutation at the yellow locus situated farther to the right in the chromosome, and in the second case the same two terminal bands were broken off spontaneously without changing the yellow locus. The present author examined the two stocks to find if those two bands are really absent or are present but do not appear. From a few slides prepared in 1941 containing the X chromosomes of the male larvae and the paired X chromosomes of the *Df(1)260-10*/Canton-S hybrid, it was found that in the paired chromosomes three configurations are present—namely, 2345/345, 2345/2345, and 12345/345—and in the unpaired male chromosomes the configuration is either 2345 or 12345. In the 2345 type of the unpaired chromosomes, the 1A2 band is often thicker and more chromatic than usual, indicating that it is possibly a compound of 1A1 and 2 bands. Actual observation of all the terminal bands in some of the single X chromosomes makes it definite that there is no true deficiency at the terminal region of the X chromosome in this stock. In the *Df(1)260-19* stock on the other hand there seems to be a genuine terminal deficiency. In the X chromosomes of both the male and female, the configuration is 345 and 345/345, respectively, in all the observed cases. In the *Df(1)260-19*/Canton-S combination the configuration is 2345/345 or 2345/5 in all the observed X chromosomes. In the male X chromosomes, the 1A3 band is always very thin and stained faintly, indicating that it is a single band. In the 2345/345 configuration the 1A3 band of the shorter chromosome is also thin and feebly chromatic, indicating that the shorter chromosome is most likely the one from the *Df(1)260-19*. If so, the X chromosome from the Canton-S is apparently so influenced by its shorter pairing partner that the 1A2 band is separated from the 1A3 band very frequently, a situation similar to that found previously in Canton-S/*yw* hybrid.

Some of these cases interpreted as pseudo-deficiencies (GOLDSCHMIDT and

KODANI 1943) might be cases of variable configuration instead of pseudo-deficiencies. At least what was interpreted as a pseudo-deficiency in the Canton-S chromosome, by these authors, is definitely a case of variable configuration.

SUMMARY

A few terminal bands in the X chromosome of the salivary gland nucleus appear to be absent in some mutant and wild stocks of *Drosophila melanogaster*. These bands are actually present in the chromosome, but are closely apposed to the more proximal band and do not appear as separate individual bands. Some or all of these bands appear as independent bands when the chromosome tip is paired. A chromosome tip may influence the tip of its pairing partner so that the hidden bands may appear. The chromosome tips of some stocks exert a greater influence of this sort on the tips of the partner chromosome than those of others. A possible mechanism for the separation of the compound bands into individual bands is suggested.

ACKNOWLEDGMENT

The author wishes to express his appreciation to PROFESSOR CURT STERN for helpful criticism and suggestions.

LITERATURE CITED

BRIDGES, C. B., 1938 A revised map of the salivary gland X chromosome. Jour. Hered. **29**: 11–13.

GOLDSCHMIDT, R., and M. KODANI, 1943 Pseudo-deficiencies and translocations of chromosome tips in Drosophila. Genetics **18**: 108–113.

SUTTON, E., 1940 Terminal deficiencies in the X chromosome of *Drosophila melanogaster*. Genetics **25**: 628–635.

REPRODUCTIVE ECONOMY IN THE CHALCIDOID WASP MELITTOBIA

R. G. SCHMIEDER AND P. W. WHITING
University of Pennsylvania, Philadelphia, Pa.

Received July 17, 1946

INTRODUCTION

MALE haploidy is characteristic of the insect order Hymenoptera, of the homopterous groups Aleurodidae and iceryine Coccidae, of the anomalous beetle Micromalthus, and possibly also of the Thysanoptera, of rotifers and of all Acarina except the suborder Mesostigmata. In a recent review of the literature (WHITING 1945) it was concluded that there had been shown no convincing evidence against the extension of the general principles of sex determination proved for Habrobracon to all other species with haploid males. However, in species naturally reproducing by close inbreeding, this would involve considerable wastage of eggs.

In Habrobracon, determination of the female sex is complementary with the female heterozygous for a pair of sex factors the members of which are separated in her sons of the two haploid genotypes. The homozygous combinations occurring in fifty percent of the fertilized eggs of inbred stocks result in sterile diploid males and excess bad eggs. In outcrosses this reproductive wastage is avoided because all fertilized eggs are female-producing. It was at first suggested that the reproductive economy of outcrosses might be brought about by differential maturation of the egg (WHITING 1935) or by multiple independently segregating factor pairs (SNELL 1935).

It was later shown (WHITING 1943) that neither nonrandomness in fertilization nor multiple factors is the true explanation for the high fecundity of outcrosses in Habrobracon, but that a series of multiple sex alleles (*xa*, *xb*, *xc*, etc.) exists so that in nature the majority of matings would involve three alleles ($xa/xb \times xc$). Natural selection would act against two-allele crosses ($xa/xb \times xa$) and favor the maintenance of several alleles in a population.

Habrobracon juglandis (Ashmead) is the only species in which multiple sex alleles have been proved, but genetic work on two other members of the genus, *Habrobracon brevicornis* Wes. (SPEICHER and SPEICHER 1940) and *Habrobracon pectinophorae* Watanabe (INABA 1939) and on the chalcidoid wasp *Pteromalus puparum* (DOZORCEVA reviewed by WHITING 1940) suggests that the same scheme may be extended to these insects.

However, in several species of invertebrates with haploid males there is normally a considerable amount of inbreeding. Because of this fact, multiple sex alleles or multiple sex factors, if they exist in such closecrossed species, would be inadequate to prevent reproductive wastage. In the cottony cushion scale *Icerya purchasi*, males are infrequent and the females have been second-

arily modified into hermaphrodites with haploid sperm produced in the hermaphrodite ovary. Self-fertilization is the rule, although the hermaphrodite may mate with males so that outcrossing is possible (HUGHES–SCHRADER 1927, 1930).

DREYFUS and BREUER (1944) have studied the chromosomes of the proctotrypoid wasp *Telenomus fariai* (Lima), a parasite of the eggs of the "barbeiro" bug Triatoma. In this wasp close inbreeding is the rule, "daughters and sons of the same mother copulate before leaving the egg shell of the host. Despite the brother-sister mating, diploid males are unknown. This difference in the behavior of Habrobracon and Telenomus is in close relation with the remarkable cytological mechanism present in the latter form." According to the authors, females are heterozygous, XY, males are haploid, X, and differential egg maturation occurs retaining X in the egg. Regular loss of a chromosome fragment takes place in spermatogenesis changing X to Y so that all sperm carry Y.

Egg counts were made, viability ratios determined, and genetic studies with mutant traits carried out neither in *Icerya purchasi* nor in *Telenomus fariai*. The possibility therefore remains that inviable sex homozygotes might be produced although diploid males were not observed. It may be mentioned here that in several stocks of Habrobracon diploid males are of very low frequency due to their extreme inviability. It is doubtful whether diploid males occurring but one percent as frequently as females would be detected without a genetic marker, unless many specimens were examined cytologically. The high diploid male stocks of Habrobracon bred in the laboratory have been derived by selection. Such selection is regarded as reverse to what occurs in nature in the evolution of male haploidy in perfecting the haploid genotype. It is, moreover, uneconomical as regards food supply for the species, so that natural selection might act to reduce diploid males in two-allele crosses for this reason also. It may well be that in many haplodiploid species, all sex homozygotes are inviable.

MATERIAL AND METHODS

Four species of the chalcidoid genus Melittobia have been bred and studied considerably (references in SCHMIEDER 1933, 1938), and all are much alike in structural features, habits, and life history. The males are eyeless and short winged and also differ from the females in several other structural details and in color. They constitute but a small proportion of the population, usually not over three percent. The females parasitize the larvae of mud-dauber wasps stinging them and ovipositing upon them. Since the parasite is very small, not over two millimeters in length, hundreds of progeny may develop on a single host larva. Among these, the few males, often only one or two, mate with the large number of females, the majority of which thus become impregnated. Active oviposition occurs only if stimulated by mating. A virgin female deposits only a very few eggs which are invariably male-producing. Thus an unmated female finding a new host, may mate with her own son, and thereupon produce an abundance of daughters.

Failure of free oviposition without mating is evidently an adaptation for conservation of the food supply which would otherwise be exhausted in the production of superfluous males. Inbreeding is the rule, son to virgin mother or, more frequently, brother to sister. Outcrosses may, however, occur among the progeny if females from different sources by chance parasitize the same host. There would seem to be little or no possibility for the male to migrate from host to host, handicapped as he is structurally and by relatively short life span.

Because Melittobia belongs to a division of the Hymenoptera distantly related to Habrobracon and because of the difference in sex ratio, habits, and life history, it seemed of interest to determine whether or not and to what extent the principles demonstrated for Habrobracon are applicable for this genus. A series of experiments was therefore planned and carried out beginning in January 1945. The technical work, including both the maintenance of the stocks and the collection of data from the experiments, has been done entirely by SCHMIEDER.

Two different highly inbred stocks of a species as yet undescribed, Melittobia species-C, were used, one in the 87th inbred generation from material collected in Wabash, Indiana, the other in the 109th inbred generation of a stock from Marlton, N. J. In deriving these stocks, a single female was isolated every generation on one host and two or three sisters on another. If progeny were secured from the single female, which was usually the case, then the next generation was continued from these. Occasionally, however, it was necessary to fall back on the "mass culture" from the sisters, and thus it would be possible that the females of the next generation might mate, not with a brother, but with a cousin. Daughters of single females would mate necessarily only with brothers and frequently there would be but a single male in the brood. Theoretically then, except for the sex alleles, the females of each of the two stocks should be homozygous and the males should be similar to each other. All fraternities within a stock should be two-allele, but the outcrosses might be of the three-allele type. It might, therefore, be possible that average number of females (♀ ♀/eggs) and female ratio (♀ ♀/♂ ♂) from the outcrosses would be double those from the closecrosses if the two stocks differed in sex alleles.

In the experiments single females were transferred to new hosts every day or two, counts were made of eggs deposited, and records were taken of adult offspring developing. Percentages of eggs producing adults should be very little lower than hatchability percentages since post natal mortality is known to be very low.

NEW DATA

Table 1 gives the summaries according to types of matings made. The term selfcross will be used in this paper as a matter of convenience to designate the cross of a haploid son back to his mother. Genetically, it is presumably similar to self-fertilization in the hermaphrodite of *Icerya purchasi* with haploid sperm. It also resembles selfing in diploid species in that fifty percent of heterozygosis is thus eliminated.

The selfcrosses include progenies of five Wabash females after each was crossed to her own son and progenies of thirteen Marlton females each of which was similarly first set unmated and then crossed to her son. Two of the close-crosses excluding selfcrosses were within the Wabash stock and eight within the Marlton stock, all pairings being with brothers except one of the latter. The outcrosses included four of Wabash females by Marlton males and four of the reciprocal. The F_2 from the outcrosses were four from daughters of Marlton females by Wabash males, crossed with their brothers (Marlton) and four from

TABLE 1

Summaries according to types of matings.

	DAYS	EGGS	♀♀	♂♂	EGGS / DAYS	♀♀ × 100 / EGGS	♂♂ × 100 / ♀♀
Self crosses* (♀♀ 23–40)	131	7,611	6,976	200	58.1	91.7	2.9
Closecrosses excluding selfcrosses (♀♀ 1–6, 41–44)	83	5,044	3,405	94	60.8	67.5	2.8
Outcrosses (♀♀ 7–14)	67	4,034	1,980	50	60.2	49.1	2.5
F_2 from outcrosses (♀♀ 15–22)	79	4,765	3,088	81	60.3	64.8	2.6
Totals including later fraternities from females 1–19	454	25,559	18,671	512	56.4	72.9	2.7

* The term selfcross is used in this paper to designate mating of a haploid son back to his mother.

daughters of Wabash females by Marlton males, crossed with their brothers (Wabash).

With the totals of table 1 there were also included progenies produced by nine of the females which were mated to various males after they had exhausted their sperm supply from their first matings. About sixty eggs are laid per day by young females, but old females tend to lay fewer, as is suggested

TABLE 2

A selection from the detailed data showing successive broods and successive fraternities produced by some of the females. The three columns of figures represent numbers of eggs laid and of adult males and adult females developing in successive broods of different fraternities and in different fraternities produced by different females or by the same female mated to different males. A brood includes the progeny from eggs laid on a single host before the female was transferred to a new host. In the complete data the parent females are numbered from 1 to 44, the parental males from 1 to 59. W indicates a parent of inbred stock from Wabash, Indiana, M from Marlton, New Jersey, M/W ♀ a first generation hybrid female from Marlton mother by Wabash father, W/M ♀ from the reciprocal. ?♂ indicates

that the source of the male was not recorded. In the detailed data the number of progeny occasionally exceeds the number of eggs recorded. This is because a few eggs were missed as they were being counted.

EGGS	♀♀	♂♂
W♀ 1×brother 1		
57	22	1
77	15	0
156	20	0
137	5	0
128	109	2
161	74	3
168	90	2
106	62	2
W♀ 2×brother 3		
46	22	0
75	16	2
161	67	3
M♀ 3×brother 4		
42	23	1
78	3	0
170	8	0
M♀ 4×brother 5		
53	38	1
91	38	1
155	62	0
119	32	0
133	134	2
118	77	2
165	109	2
110	67	3
W♀ 7×M♂ 9		
56	12	0
104	34	0
132	46	1
106	27	1
124	92	3
96	92	2
W♀ 7×M♂ 10		
47	37	2
90	68	2
W♀ 7×M♂ 11		
86	64	5
137	125	3
43	18	2
70	29	3
82	49	1

EGGS	♀♀	♂♂
W♀ 8×M♂ 12		
36	35	0
160	72	4
117	22	0
120	99	2
95	79	3
W♀ 8×M♂ 13 total 9 broods		
862	709	16
W♀ 9×M♂ 14		
5	2	0
92	53	0
164	91	2
132	24	0
124	93	2
65	51	4
W♀ 9×?♂ 15 total 4 broods		
433	396	10
W♀ 9×?♂ 16 total 3 broods		
237	192	7
W♀ 10×M♂ 17		
8	3	0
114	42	2
140	84	3
111	40	1
101	92	2
117	108	2
57	52	2
39	34	0
2	0	0
W♀ 10×M♂ 18 total 6 broods		
788	712	18
M♀ 11×W♂ 19		
16	4	0
110	8	0
129	59	1
121	38	0
112	91	2
6	3	2

EGGS	♀♀	♂♂
M♀ 12×W♂ 20		
73	17	1
152	36	0
161	106	2
145	65	2
107	111	2
4	0	2
M♀ 12×M♂ 21 total 3 broods		
143	111	2
M♀ 12×M♂ 22		
146	77	2
M♀ 12×W♂ 23		
137	49	1
128	120	1
107	88	3
M♀ 12×son 24		
115	67	1
40	21	1
15	9	1
76	58	1
54	51	2
10	7	1
M♀ 13×W♂ 25		
40	5	2
124	1	0
141	36	0
115	13	1
31	25	4
M♀ 13×M♂ 26		
38	21	0
65	58	0
M/W♀ 15× brother 28		
101	50	1
106	90	3
153	135	4
141	51	0
152	144	6
2	0	0
M/W♀ 15×M♂ 29		
82	53	1

EGGS	♀♀	♂♂
M/W♀ 16× brother 30		
58	46	1
75	67	2
115	107	2
104	59	2
119	111	2
105	85	2
52	9	0
M/W♀ 17× brother 31		
48	33	0
76	56	1
133	127	1
W/M♀ 19× brother 33 total 7 broods		
943	659	22
W/M♀ 19×?♂ 34		
19	14	0
67	4	0
W/M♀ 20× brother 35		
126	82	2
120	79	1
103	83	3
128	27	1
168	43	1
132	76	1
111	74	2
49	46	2
51	25	0
84	2	0
107	59	1
45	26	2
W/M♀ 21× brother 36		
173	110	2
134	96	3
134	123	2
138	69	1
200	72	3
99	56	2

in the selfcrosses and in the later crosses. Ratio of males to females proved to be somewhat less than three percent. It appears to fluctuate very little in the different groups of crosses shown in table 1 or even in the detailed data shown in table 2.

High fecundity of closecrosses

As regards eggs developing into females, however, there are wide differences. Data are inconsistent with expectation based on results from Habrobracon. Eggs from the closecrosses produce more females than those from the outcrosses instead of the reverse, and in the selfcrosses with more than ninety percent of the eggs producing adult females there is no possibility of an equal number of "bad" eggs which might correspond to the homozygous diploid male-producing combinations expected in two-allele crosses. F_2 progenies from the outcrosses closely approximate the offspring from the closecrosses. The data suggest that, on the average, the more closely related the parents, the higher the fertility of the eggs.

Fluctuating and uniform fraternities

The detailed data reveal some interesting conditions which cannot be brought out by the summaries. Hence table 2 has been prepared, representing a selection from these data and showing the different broods of some of the fraternities and also the different fraternities derived by different matings of some of the females. A brood is here defined as the progeny produced on a single host and thus developing from the eggs laid over a period of one or two days according as the mother was transferred from host to host. After a varying number of transfers a female would cease laying eggs, thus indicating exhaustion of her sperm supply. She might then be mated to a second male to produce a second fraternity.

The single fraternity produced by W ♀ 1 crossed with her brother consisted of eight different broods. Numbers of eggs laid for the first two broods were smaller than those from the later merely because transfers were made sooner. Offspring produced per egg fluctuate very widely, being especially low in the third and fourth broods but increasing suddenly in the fifth. After the eighth transfer only one egg was laid, indicating exhaustion of sperm supply.

The outcross of W ♀ 7 by M ♂ 9 resulted in six broods showing wide deviations in adults produced. After the female ceased ovipositing she was outcrossed a second and again a third time to Marlton males 10 and 11, respectively.

If there was very little deviation in viability among the broods of a fraternity, only the totals are given. Thus W ♀ 8, giving a fluctuating fraternity by M ♂ 12, gave a uniform fraternity by M ♂ 13 and W ♀ 9, after producing a fluctuating fraternity by M ♂ 14, gave two uniform fraternities by males of unrecorded origin. Some fraternities consist of but a single brood as the third produced by M ♀ 12. Her second was unform and her fifth, from a selfcross by son 24, was fluctuating, viability being about fifty percent in the first three broods but rising to near 100 percent in the fifth. The second brood

from M♀ 13 outcrossed to W♂ 25 resulted in but a single adult from 124 eggs, while the fifth gave 29 from 31 eggs. It is unnecessary to comment further on the selected data which may be studied in table 2.

Since broods from the selfcrosses of females 23–40 (table 1) were, in general, very uniform, showing insignificant deviation from the high average 91.6 percent, the detailed data are omitted from table 2. A selection of the more divergent broods showed four with females per egg about 71 percent (194 females from 271 eggs) and in one small brood there were only 29 females from 58 eggs.

The data suggest that the difference between fluctuating and uniform fraternities is genetic. The broods of the uniform fraternities are all of high viability, no fraternities being as yet obtained with broods of uniformly low viability.

The cause of the differences between broods within a fluctuating fraternity is unknown. An attempt was made to determine whether or not food might influence viability. Larvae of mud-dauber wasps may be collected in early spring and refrigerated until it is desired to use them. After several months they lose their capacity to metamorphose, but they serve well as Melittobia food for several years. Of ten Marlton females eclosing in March 1946, nine were mated with their own sons and one was mated with a Marlton male from another culture. Five had been fed before mating on *Trypoxylon politum* larvae collected in March 1945, and four had been fed on *Sceliphron caementarius* larvae collected in March 1942. After mating, some of the females were fed on Trypoxylon collected in 1943, 1945, and 1946 and Sceliphron collected in 1942 and 1946. The brood showing the lowest hatchability, 29 females and one male from 58 eggs, was 1942–Sceliphron fed, and one fraternity of eight broods totaling 686 females and 20 males from 744 eggs was 1946–Sceliphron fed. In general there was little association, however, between hatchability and type of host used but the experiment was carried out with crosses, mostly selfcrosses, giving relatively uniform fraternities of high viability.

As a further check on the possibility that condition of the host might affect hatchability, three sisters (M♀ ♀ 42–44) were selected from one of the selfcross fraternities (M♀ 32×son 47) and crossed with their brothers (M♂ ♂ 57–59). Females 42 and 43 were fed and their offspring reared on Sceliphron hosts collected in March 1943, the poorest, discolored, shrunken host larvae available being used. For female 44, relatively fresh *Trypoxylon clavatum* was used, mostly 1946 material. From the two "poorly fed" cultures there were produced 567 females and 16 males from 603 eggs and 201 females and five males from 215 eggs. From the well-fed culture there developed 504 females and 16 males from 538 eggs. Viability was uniformly high in the different broods of the three fraternities despite difference in feeding.

Offspring of unmated females

For the selfcrosses in the feeding experiment 11 Marlton stock females had been set unmated in March, 1946. The six that were fed *Trypoxylon politum* larvae collected in March 1945 laid 15 eggs, all of which developed into adult

males. The five that were fed *Sceliphron caementarius* larvae collected in March 1942 laid 14 eggs, only six of which were viable. There is here a suggestion that viability of unfertilized eggs may be affected by age or by species of the host.

Only 29 eggs were produced in 120 days, the total active life of the 11 females before they were mated. This rate of 0.24 egg per day stands in marked contrast to the rate after mating, almost 60 per day.

Relative viability of males and females

Table 1 indicates no difference in sex ratio associated with the different types of matings, suggesting that viability of the males is positively correlated with that of the females. As a further check on this point the data from the fluctuating fraternities were analyzed according to broods. The broods were grouped according to percentage of eggs producing females. Seventeen broods of very low female production (1–20 percent) totalled 1,571 eggs; 21 of low female production (21–40 percent) totalled 2,174 eggs; 35 of intermediate (41–60 percent) totalled 3,616 eggs; 35 of high (61–80 percent) totalled 3,482 eggs; and 53 of very high (81–100 prcent) totalled 5,376 eggs. For the five groups the numbers of males were, respectively, 5, 15, 61, 66, 121, totalling 268. Percentages of eggs producing males were therefore, respectively, 0.3, 0.7, 1.7, 2.0, and 2.25. Production of males is positively correlated with production of females. A χ^2 test indicated no significant deviation in slope between the two distributions.

GENERAL DISCUSSION

Among closecrossed animals with haploid males, Melittobia is advantageous in that it is possible to count the eggs and subsequently make record of the progeny. In Telenomous and other egg parasites in which several individuals develop within a single host egg and mating occurs before emergence, it maybe argued that the Habrobracon scheme applies with wastage of 50 percent of the zygotes. Since it has now been established that this cannot be the case, in Melittobia at least, we may look for some adaptive mechanism associated with sex determination which has been developed for reproductive economy in other closecrossed species. Multiple allelism may be the more primitive and general method functioning for outcrossed species, with the closecrossed species adopting various modifications, but retaining female heterozygosis.

The data presented in this paper suggest that in Melittobia the closer the mating the higher the fecundity. If some type of non-randomness in fertilization has been developed for economy in inbreeding, it is possible that crossing of different stocks may so upset the balance that the mechanism fails to function perfectly, resulting in infertile eggs, homozygotes. The data are not yet adequate to establish that outcrosses are, in general, less fecund than closecrosses. Differences between the fluctuating and the uniform fraternities may be due to some untested environmental factor.

No diploid males have yet been found in Melittobia, all males studied cytologically having five chromosomes. Some of the infertile eggs produced with

the fluctuating fraternities or even some of the few with the uniform fraternities may possibly represent sexhomozygotes.

The questions raised here may in time be answered if a sex linked mutant gene can be obtained.

SUMMARY

In an undescribed species of the chalcidoid genus Melittobia close inbreeding is the rule, but outcrossing may occur. Unmated females lay very few eggs, and these are male producing. After mating occurs, an abundance of eggs is laid which, for the most part, develop into females, males constituting less than three percent of the offspring.

The experiments including egg counts show that despite close inbreeding with "selfcrosses" of haploid sons back to mothers, there are no diploid males and no "bad" eggs as in Habrobracon sex homozygotes. Moreover, outcrosses have been less fecund than closecrosses. The methods of reproductive economy and of sex determination have not been determined.

Fraternities fall into two groups as regards successive broods on different hosts—the uniform with broods of high fecundity and the fluctuating with some broods of very low fecundity. These differences appear not to be caused by food but may be genetic.

It is suggested that in haplodiploid species multiple sex allelism may be the more primitive and general method of reproductive economy and that the closecrossed species have adopted some other method.

LITERATURE CITED

DREYFUS, A., and M. E. BREUER, 1944 Chromosomes and sex determination in the parasitic hymenopteron *Telenomus fariai* (Lima). Genetics **29**: 75–82.

HUGHES-SCHRADER, S., 1927 Origin and differentiation of the male and female germ cells in the hermaphrodite of *Icerya purchasi* (Coccidae). Z. Zellforsch. **6**: 509–540.

1930 The cytology of several species of iceryine coccids with special reference to parthenogenesis and haploidy. J. Morph. Physiol. **50**: 475–495.

INABA, FUMIE, 1939 Diploid males and triploid females of the parasitic wasp *Habrobracon pectinophorae* Watanabe. Cytologia **9**: 517–523.

SCHMIEDER, R. G., 1933 The polymorphic forms of *Melittobia chalybii* Ashmead and the determining factors involved in their production (Hymenoptera: Chalcidoidea, Eulophidae). Biol. Bull. **65**: 338–354.

1938 The sex ratio in *Melittobia chalybii* Ashmead, gametogenesis and cleavage in females and in haploid males (Hymenoptera: Chalcidoidea). Biol. Bull. **74**: 256–266.

SNELL, GEORGE D., 1935 The determination of sex in Habrobracon. Proc. Nat. Acad. Sci. **21**: 446–453.

SPEICHER, B. R., AND KATHRYN G. SPEICHER, 1940 The occurrence of diploid males in *Habrobracon brevicornis*. Amer. Nat. **74**: 379–382.

WHITING, P. W., 1935 Sex determination in bees and wasps. J. Hered. **26**: 263–278.

1940 Sex-linkage in Pteromalus. Amer. Nat. **74**: 377–379.

1943 Multiple alleles in complementary sex determination of Habrobracon. Genetics **28**: 365–382.

1945 The evolution of male haploidy. Quart. Rev. Biol. **20**: 231–260.

FURTHER STUDIES ON THE GENETICS OF THALASSEMIA

JAMES V. NEEL[1] AND WILLIAM N. VALENTINE[2]

The Department of Medicine, The University of Rochester School of Medicine and Dentistry and the Medical Clinic of the Strong Memorial and Rochester Municipal Hospital, Rochester, New York

Received August 16, 1946

INTRODUCTION

In 1925 COOLEY and LEE for the first time clearly defined as a distinct clinical entity a chronic, progressive, and fatal anemia of childhood which is characterized hematologically by hypochromia, micro-, aniso-, and poikilocytosis, decreased erythrocyte fragility in hypotonic saline solutions, peripheral erythroblastosis, failure to respond to any known anti-anemic therapy, and an increased incidence of oval and "target" red blood cells. These latter are erythrocytes which on stained smears instead of the usual achromic center appear to have a central dot and peripheral ring of hemoglobin separated by an intervening zone of pallor. Some fifteen years later WINTROBE, MATHEWS, POLLACK and DOBYNS (1940), DAMESHEK (1940), and STRAUSS, DALAND, and FOX (1941) independently described an anemia qualitatively similar to that described by COOLEY and LEE but quantitatively very much less severe, never terminating fatally. Both anemias were found to be familial, and confined in large part to the socalled Mediterranean peoples—Italians, Greeks, Turks, Cypriots, etc., and their descendants. Although ANGELINI (1937, quoted from WINTROBE *et al.*, 1940) and CAMINOPETROS (1938) had observed fragility changes in the blood of parents of patients with the severe anemia described by COOLEY and LEE, it remained for WINTROBE (1942) and SMITH (1942) to point out that the above mentioned mild anemia was present in the parents and some of the siblings of children with COOLEY'S anemia. This finding has been confirmed by DAMESHEK (1943), SMITH (1943), and VALENTINE and NEEL (1944).

Three theories regarding the inheritance and relationship of these two anemias have been advanced, namely, (1) that the severe disease is due to homozygosity for an inherited factor which when heterozygous produces the mild condition, an hypothesis adumbrated by CAMINOPETROS (1938), and advanced later by DAMESHEK (1943), (2) that both anemias are due to a variably expressed dominant (SMITH, 1943), and (3) that the severe anemia is due to the interaction of two non-allelic dominant factors (MCINTOSH and WOOD, 1942). This latter theory was presented at a time when the only recognized abnormality in the blood of parents of patients with the severe disease was the fragility change (CAMINOPETROS, 1938); it was suggested that

[1] Now First Lieutenant, M.C., A.U.S.

[2] Now Captain, M.C., A.U.S.

one of the two dominants postulated might be responsible for these fragility changes. Elsewhere we (1944) have reviewed these three theories in the light

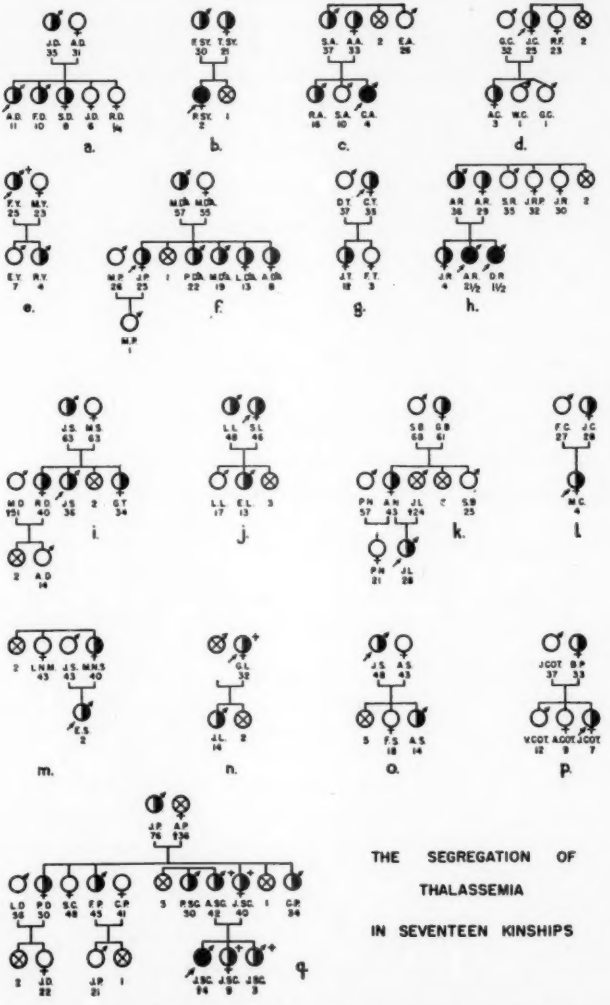


FIGURE 1.—The segregation of thalassemia in 17 kinships. Solid circles indicate thalassemia major. Half solid circles—thalassemia minor. Open circles—normal individuals. Open circles containing an X indicate individuals not seen, the number below the circle showing the number of persons involved. → points to the proband. + signifies that the data on this individual have been published in VALENTINE and NEEL (1944).

of the available data and have found that the first and the third theories come closer to satisfying all of the facts. Theory (1) was adopted as a working hypothesis because of its greater simplicity. It was suggested that the two anemias be designated, in view of their probable homozygous-heterozygous relationship, as thalassemia major and thalassemia minor, the term thalassemia having been applied to the severe disease by WHIPPLE and BRADFORD (1936). The frequency of thalassemia major was found to be one in each 2,368 births in the Italian element of the population of Rochester, N. Y., from which,

TABLE 1

Hematological findings in the DE M. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
A. DEM., proband	11	10.2	5.715	31.0	54.2	17.8	32.9	3+	2+	0.44	0.22
										*c0.48	0.36
J. DEM., father	35	13.7	6.43	42.6	66.2	21.2	32.1	3+	1+	0.44	0.24
										*c0.48	0.36
A. DEM., mother	31	13.7	4.83	41.0	84.8	28.3	33.4	0	0	0.48	0.32
										*c0.48	0.36
F. DEM., brother	10	11.6	5.71	37.0	64.8	20.3	31.4	2+	2+	0.46	0.22
										*c0.48	0.32
S. DEM. sister	8	11.6	6.87	37.5	54.6	16.9	30.9	3+	3+	0.42	0.20
										*c0.48	0.32
J. DEM., sister	6	13.4	4.78	39.5	82.6	28.0	33.9	0	1+	0.46	0.30
										*c0.48	0.32
R. DEM., sister	½	13.1	4.64	35.9	77.3	28.2	36.4	0	0	0.48	0.30
										*c0.48	0.32

* c signifies control values.

assuming the homozygous-heterozygous relationship, the frequency of thalassemia minor was calculated to be about one in 25 (NEEL and VALENTINE, 1945).

In our analysis of the 13 reported marriages between a normal individual and one with thalassemia minor there appeared to be among the offspring a significant excess of children with thalassemia minor over the expected 1:1 ratio. No such tendency towards an excess of thalassemia minor was apparent in the 13 reported marriages between two individuals with thalassemia minor, where the segregation of the offspring into three groups: normal, thalassemia minor, and thalassemia major, was studied. Here the ratio of thalassemia minor to normal showed a good approximation to the expected 2:1 ratio on the basis of a small sample. This unexplained discrepancy was deemed worthy of further investigation. It is the purpose of the present paper to present hematological data on 17 additional kinships in which the inheritance of thalassemia major or minor can be studied, with an analysis of the observed ratios, after which certain problems of population dynamics will be considered.

METHODS

The kinships comprising this study, with one exception, came to our attention when a member of the family was seen in STRONG MEMORIAL or ROCHESTER MUNICIPAL HOSPITAL and found to have either thalassemia major or minor. The one exception was a family being followed at the GENESEE HOSPITAL, two members of which had thalassemia major. Individuals with thalassemia major in all instances presented with symptoms referable to the anemia. Individuals with thalassemia minor, on the other hand, presented with a variety of complaints, and the anemia was for the most part an incidental finding.

TABLE 2

Hematological findings in the SY. *1 kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
P.Sy.,* proband	2	5.6	2.615	17.0	65.0	21.2	32.6	2+	2+	0.48	0.20
										co.48	0.32
F.Sy., father	30	13.3	7.11	42.0	59.0	18.7	31.7	3+	2+	0.42	0.20
										co.48	0.32
T.Sy., mother	21	10.4	5.26	34.3	65.2	19.7	30.3	0	1+#	0.44	0.26
										co.48	0.32

* Studies carried out after the patient had received one transfusion.

Despite the scarcity of target and oval cells, the smear showed quite significant micro- and anisocytosis, and hypochromia.

The diagnosis of thalassemia in any individual rested on the following considerations:

(1) *The erythrocyte constants.* Each of the individuals included in this report had, with very few exceptions, two or more erythrocyte counts and hemoglobin determinations, and a single hematocrit determination. From these were calculated the mean corpuscular volume (MCV) in cubic microns, the mean corpuscular hemoglobin content (MCH) in micro-micrograms, and the percent of the erythrocyte, composed of hemoglobin (MCHC), these being cellular constants in terms of which it is readily possible to compare individuals. The usually accepted normal range of values for the MCV is stated by WINTROBE (1942) to be 82–92 cubic microns. The normal MCH ranges from 27 to 31 micro-micrograms. The normal MCHC is 32–36%. These values are based on WINTROBE'S extensive personal experience and a compilation of the literature, and are given as the range within which 93% of healthy men and women fall. However, when one examines some of the publications in which constants are stated in terms of mean and standard deviation, one finds, for example, an MCV of 85.9 ± 5.1 (FOSTER and JOHNSON, 1931), 89.2 ± 3.6 (ANDRESEN and MUGRAGE, 1936), and 86.5 ± 7.9 (HAMRE and AU, 1942). This would indicate that an appreciable number of individuals would show

MCV's in the 70–80 range. In the above quoted series, other erythrocyte constants also tend to have a wider distribution than WINTROBE has indicated. The values for newborn infants tend to be somewhat higher than for adults, while for children between the ages of three months and adolescence they tend to be somewhat lower. It is characteristic of thalassemia that the MCV, MCH, and MCHC are decreased.

(2) *The appearance of a stained blood smear.* In addition to an examination for hypochromia, anisocytosis, and poikilocytosis, particular attention was directed towards the occurrence of abnormal numbers of oval and "target" red blood cells, these being cell types especially characteristic of (though not

TABLE 3

Hematological findings in the A. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
C.A., proband	4	7.4	3.72	25.6	69.0	19.4	28.9	4+	2+	0.38 CO.42	0.20 0.32
S.A., SR., father	37	9.6	4.12	30.3	73.5	23.3	31.6	1+	1+	0.44 CO.48	0.26 0.34
A.A., mother	33	12.3	5.8	37.5	64.6	21.2	32.8	0	2+	0.48 CO.48	0.30 0.34
R.A., brother	16	13.4	6.315	41.7*	66.0	21.2	32.1	1+	1+	— —	— —
S.A., brother	10	13.5	4.39	38.0	86.5	30.8	35.6	0	0	0.48 CO.50	0.32 0.32
E.A., uncle	26	16.7	5.49	45.8	83.4	30.4	36.4	0	0	0.50 CO.52	0.34 0.30

confined to) thalassemia. Slides were graded from 0 to 4 with respect to these two cell types, slides graded as 0 showing only the normal number of such cells, slides graded as 4 showing greatly increased numbers. An occasional normal person may show 1+ ovalocytes or target cells.

(3) *The fragility of the erythrocytes as measured by resistance to hypotonic saline solutions.* Cell fragility is characteristically decreased in thalassemia.

(4) *A familial incidence of a similar blood picture.*

(5) *Lack of evidence of iron deficiency, chronic blood loss, or other disease which might so distort the blood findings as to produce a picture comparable to thalassemia,* or else reasonable evidence that when other disease does coexist, it is not responsible for the hematological findings.

(6) *Failure to respond to any known therapy.*

DATA

Each of the families studied is briefly described below. A tabular summary is presented of the hematological findings on each kinship, and the segregation of individuals as interpreted from the data is indicated in figure 1. As will be

apparent, the segregation is for the most part clear cut. The possibility of misclassification will be discussed later. All of the individuals studied, so far as

TABLE 4

Hematological findings in the Co. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
J.Co., proband	25	11.1	6.795	34.3	50.5	16.3	32.3	4+	3+	0.46	0.20
										co.48	0.34
(2.8.44)										0.46	0.22
(3.25.46)	27	11.5	6.40	37.0	57.8	18.0	31.1	4+	2+	co.50	0.34
										0.48	0.30
G.Co., SR., husband	32	14.9	4.93	42.8	87.3	30.4	34.8	0	0	co.50	0.32
										0.44	0.20
A.Co., daughter	3	10.3	5.925	35.0	59.0	17.4	29.4	2+	2+	co.48	0.34
W.Co., son											
(2.8.44)	1	9.9	5.415	34.8	64.2	18.3	28.5	1+	1+	0.50	0.24
										co.48	0.34
(3.25.46)	3	13.6	4.37	38.5	88.0	31.3	35.3	0	0	0.44	0.32
										co.50	0.34
G.Co., JR., son											
(2.8.44)	1	10.35	5.125	37.0	72.2	20.1	27.8	1+	1+	0.50	0.24
										co.48	0.34
(3.25.46)	3	13.4	4.36	38.0	87.2	31.4	35.3	0	0	0.46	0.22
										co.50	0.34
R.F., sister	22	12.7	4.355	36.3	83.2	29.2	35.0	0	0	0.48	0.32
										co.48	0.34

TABLE 5

Hematological findings in the Y. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
M.Y., wife	23	14.6	4.335	41.5	95.6	33.6	35.1	0	0	0.50	0.32
										co.52	0.34
E.Y., son	7	13.0	4.06	37.3	91.8	32.0	34.8	0	0	0.48	0.32
										co.52	0.34
R.Y., son	4	10.7	5.85	36.5	62.4	18.3	29.3	4+	1+	0.42	0.20
										co.48	0.30

could be determined, were of Southern Italian and Sicilian extraction. As a rule, the cooperation obtained from this group was poor, and this fact, together with the war and migration, accounts for many of the obvious gaps in pedigrees.

(1) *The D. kinship.* The proband, A.D., #59430, an 11 year old male, was hospitalized because of a swollen, painful right knee. A diagnosis of acute rheumatic fever was reached. Thalassemia minor was an incidental finding. The results of studies on the patient, his parents, and four sibs are shown in table 1 and figure 1a. HUBBARD and McKEE (1939) have shown that exacerbations of rheumatic fever may be accompanied by a mild to moderately severe anemia. This fact may influence the details of the picture in the proband, but the appearance of his erythrocytes plus the existence of a similar condition in the father and two sibs leave little doubt that he actually had thalassemia minor

TABLE 6

Hematological findings in the D'A *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
J.D'A. P., proband	25	11.7	5.845	39.3	67.2	20.0	29.8	3+	3+	0.46 co.48	0.20 0.32
M.D'A., SR., father	57	13.3	6.53	42.3	64.9	20.4	31.4	3+	1+	0.46 co.50	0.24 0.32
M.D'A., mother	55	14.6	4.405	43.0	97.6	33.2	34.0	0	0	0.48 co.50	0.30 0.32
P.D'A., brother	22	14.9	6.34	44.0	69.5	23.5	33.8	1+	1+	0.48 co.50	0.26 0.32
M.D'A., JR., brother	19	14.1	5.615	45.1	80.3	25.1	31.2	2+	1+	0.44 co.50	0.24 0.32
L.D'A., sister	13	13.9	5.70	40.8	71.5	24.4	34.0	1+	1+	0.44 co.50	0.24 0.32
A.D'A., sister	8	11.4	6.095	36.8	60.4	18.7	31.0	3+	2+	0.44 co.50	0.20 0.32
M.P., SR., husband	26	16.2	5.69	46.3	81.4	28.5	35.0	0	0	0.46 co.50	0.32 0.32
M.P., JR., son	1	12.7	5.315	40.4	75.4	23.9	31.4	1+	0	0.48 co.50	0.28 0.32

(2) *The Sy 1 kinship.* The proband, P.SY., #209842, was a two year old girl admitted to the hospital because of anorexia, listlessness, and retarded development. She was found to have thalassemia major, and was discharged after a brief hospital stay, somewhat improved as a result of transfusion therapy. Her second and last admission was one month after the first, and was prompted by a rapidly developing, nonthrombocytopenic purpura which proved fatal on the ninth hospital day. The results of studies on the patient and her parents are shown in table 2 and figure 1b.

(3) *The A. kinship.* The proband, C.A., #212689, a four year old male, was referred to this clinic at age two years because of retarded development, anemia, and a palpable spleen. A diagnosis of thalassemia major was reached, and the child discharged after transfusion therapy. There have been four admissions in the two years since the first one, usually because of an upper respira-

tory infection and symptoms referable to the anemia, and on each occasion the child has required transfusion. The results of studies on the patient, his parents, two brothers, and one uncle are given in table 3 and figure 1c. Two months after our studies the patient's father entered the hospital complaining

TABLE 7

Hematological findings in the T. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
C.T., proband	35	11.6	5.79	36.3	62.7	20.0	32.0	2+	2+	0.48 co.50	0.22 0.34
D.T., husband	37	17.5	5.875	49.0	83.5	29.8	35.8	0	0	0.52 co.50	0.36 0.34
J.T., daughter	12	12.0	5.80	36.0	62.0	20.7	33.3	1+	3+	0.44 co.50	0.22 0.34
F.T., daughter	3	12.6	4.115	36.8	89.4	30.6	34.2	0	0	0.50 co.50	0.32 0.34

TABLE 8

Hematological findings in the R. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
A.R., JR.,* proband	1½	8.6	3.02	22.1	73.2	28.4	38.9	1+	1+	0.46 co.48	0.24 0.36
A.R., SR., father	36	14.2	5.85	42.0	71.8	24.3	33.8	3+	2+	0.46 co.48	0.24 0.36
A.R., mother	29	11.7	5.10	34.0	66.6	22.9	34.4	4+	1+	0.44 co.48	0.20 0.36
J.R., sister	4	11.7	5.98	34.0	56.8	19.6	34.4	3+	2+	0.42 co.48	0.22 0.36
S.R., uncle	35	16.8	5.565	47.5	85.3	30.2	36.4	0	0	0.46 co.48	0.30 0.34
J.R.P., aunt	32	14.6	5.01	40.7	81.2	29.1	35.9	0	1+	0.48 co.48	0.30 0.34
J.R., aunt	30	13.1	4.57	39.0	85.3	28.7	33.6	0	0	0.46 co.48	0.32 0.34

* Transfused one week before study.

of severe, bloody diarrhea and was found to have multiple polyps of the colon with malignant degeneration and metastases to the liver, from which he expired two years later. It seems probable that early malignant changes were present in the bowel at the time of our studies, and may have contributed to the degree of anemia observed (hemoglobin 9.6 gms. percent).

(4) *The Co. kinship.* The proband, J.Co., #214598, age 25, was referred to the hospital for the study of a mild anemia first discovered four years previously, when she underwent an appendectomy. It had proved resistant to various therapeutic measures. In addition, the patient at the time of admission had symptoms typical of a mild situational depression, precipitated by domestic difficulties. The initial findings on the patient, her husband, three children,

TABLE 9

Hematological findings in the S. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS BEGINS	HEMOLYSIS COMP.
J.S., proband	36	13.05	5.635	39.0	69.3	23.2	33.5	0	2+	0.48	0.26
										co.48	0.32
J.S., father	63	12.0	5.445	36.2	66.5	22.1	33.2	2+	1+	0.46	0.24
										co.50	0.34
M.S., mother	63	14.8	5.525	43.0	78.0	26.8	34.4	0	0	0.48	0.32
										co.50	0.34
R.D., sister	40	13.2	5.93	37.5	63.[illegible]	22.2	35.2	1+	1+	0.46	0.24
										co.48	0.30
A.D., nephew	14	13.2	4.20	—	—	31.4	—	0	0	—	—
G.T., sister	24	12.2	5.705	39.0	68.3	21.4	31.2	3+	2+	0.46	0.24
										co.50	0.34

TABLE 10

Hematological findings in the LO. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS BEGINS	HEMOLYSIS COMP.
S.L., proband	46	12.3	5.125	36.8	71.8	24.0	33.4	1+	1+	0.46	0.26
										co.50	0.36
L.L., SR., husband	47	15.0	5.715	45.5	79.5	26.2	33.0	2+	1+	0.48	0.26
										co.50	0.36
L.L., JR., son	15	15.3	5.565	45.0	80.8	27.5	34.0	0	1+	0.48	0.30
										co.50	0.36
E.L., son	10	12.9	6.21	38.0	61.1	20.7	33.9	1+	2+	0.44	0.24
										co.50	0.36

and a sister, are given in table 4 and fig. 1d. The twins were identical and were thought to be carried to term. The initial observations on the twins disclosed an abnormal blood picture, compatible with thalassemia minor. However, it is known that twins and premature infants are especially subject to the microcytic, hypochromic anemias of infancy. Moreover, it seemed strange that there should be differences of the magnitude observed in the blood of identical

twins. Accordingly, the twins and MRS. Co. were restudied some two years after the initial observations. At this time the blood of the twins was normal, while the findings on MRS. Co. were much as before.

(5) *The Y. kinship.* The proband, F.Y., a 25 year old white male, was the F.Y. of fig. 1 and table 1 in VALENTINE and NEEL (1944). He came to our attention at that time as the uncle of a child with thalassemia major, and was himself found to have thalassemia minor. Since the publication of that paper it has been possible to study this man's wife and two children (table 5 and fig. 1e).

TABLE 11

Hematological findings in the L. *kinship*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
J.L., proband	28	12.15	5.51	37.0	67.2	22.1	32.8	4+	1+	0.42	0.20
										co.48	0.34
A.N., mother	43	11.9	5.47	34.7	63.2	21.7	34.3	2+	2+	0.44	0.24
										co.48	0.34
P.N., step-fa.	57	15.2	5.49	43.3	78.8	27.7	35.1	0	0	0.48	0.30
										co.48	0.30
P.N., step-si.	21	13.2	4.63	38.0	82.1	28.5	34.7	0	0	0.46	0.30
										co.48	0.30
S.B., grand-fa.	68	13.7	4.335	42.6	98.2	31.6	32.1	0	0	0.50	0.30
										co.50	0.34
G.B., grand-mo.	61	12.0	5.33	37.5	70.4	22.5	32.0	2+	2+	0.46	0.24
										co.50	0.34
J.B., uncle	25	16.7	4.995	47.0	94.0	33.4	35.5	0	0	0.50	0.34
										co.50	0.34

(6) *The D'A kinship.* The proband, J.D'A.P., #11484, was a 25 year old white married woman found at age 15 to have a mild anemia which could not be accounted for on the basis of organic disease, and which persisted despite extensive iron therapy and what appeared to be an adequate diet. The results of hematological studies on the patient, her husband and son, and her parents and four siblings are shown in table 6 and figure 1f.

(7) *The T. kinship.* The proband, C.T., was a 35 year old white woman who had been treated with iron and liver preparations for several years by a local physician because of a low hemoglobin. She was referred to our attention after studies on another of this physician's patients (J.Co.) with a similar history had disclosed thalassemia minor. The findings on the proband, her husband, and two children are given in table 7 and figure. 1g.

(8) *The R. kinship.* The probands of this family, A.R., JR., and D.R. boys of 2½ and 1½ years of age respectively, were being followed at the GENESEE HOSPITAL with a diagnosis of thalassemia major at the time they came to our attention. A.R. had been first seen there at age 14 months, with complaints

of sallowness and growth retardation, and a diagnosis of thalassemia major made. D.R., his younger brother, was seen at age 13 months with similar complaints and a like diagnosis. Both had received transfusion therapy, A.R. just a week before these studies. In table 8 and figure 1h are given the results on A.R., his sister, his parents, and a paternal uncle and two aunts. The examina-

TABLE 12

Hematological findings in the C. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
M.C., proband	4	13.1	5.73	38.0	66.3	22.8	34.5	1+	2+	0.44 co.46	0.22 0.32
F.C., father	27	17.6	5.045	45.0	89.2	34.9	39.1	1+	0	0.48 co.48	0.30 0.32
J.C., mother	28	13.1	5.295	40.0	75.6	24.7	32.8	3+	3+	0.44 co.48	0.22 0.32

TABLE 13

Hematological findings in the SV. 2 *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
E.S., proband	2	12.8	5.895	39.1	65.7	21.7	32.7	3+	1+	0.46 co.56	0.22 0.38*
J.S., father	43	17.4	5.77	50.5	87.4	30.3	34.4	0	0	0.56 co.56	0.28 0.34
M.N.S., mother	40	11.1	6.085	39.8	65.4	18.2	27.9	2+	2+	0.48 co.56	0.26 0.34
L.N.M., aunt	43	14.4	4.435	41.5	93.6	32.5	34.7	1+	0	0.50 co.54	0.32 0.30

* The control fragility values on the first three members of this kinship are high. This can be attributed to contamination of the test solutions, which were restandardized as soon as this was observed.

tion of D.R. was limited to a blood smear, which was typical of thalassemia major. The findings on A.R., JR., were atypical, probably because he had been transfused so recently and, from the appearance of his smear, only a minority of the erythrocytes in circulation were actually his own.

(9) *The S. kinship.* The proband, J.S., #63819, was a 35 year old married man who entered the hospital for study of a chronic, low grade anemia. He had been subject to recurrent episodes of fatigue all his life, and six years prior to admission was told he was anemic. Since that time he had received con-

siderable iron and liver therapy, but the anemia had persisted. The findings on the patient, his parents, two sisters, a brother-in-law, and a nephew are entered in table 9 and figure 11. Both of the affected sisters had been previously told they were anemic, and were willing to cooperate in family studies. However, the patient's two siblings who were not studied had never been told they were anemic, and, because of family antagonisms, were unwilling to come to the hospital. There is the possibility here, then, of a definite sampling error, and this family is excluded from later calculations. The patient's brother-in-law, M.D., died in this hospital at age 51 of multiple myeloma,

TABLE 14

Hematological findings in the LA. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
J.La., son	14	10.8	5.69	34.5	60.6	19.0	31.6	4+	2+	0.40 CO.48	0.20 0.32

two years prior to these studies. Studies carried out during his fatal illness make it very unlikely he had thalassemia minor. Because of poor cooperation the patient's nephew was only partially studied, but such examinations as were performed revealed no abnormalities.

(10) *The Lo. kinship.* The proband, S.L., #239348, was a 46 year old white married woman who some six months prior to entry had developed weakness and fatigue. The physician whom she consulted found her hemoglobin to be low, and prescribed iron and liver therapy, without effect. She was therefore referred here for further study. Table 10 and figure 1j contain the results of the examination of the patient, her husband, and two of her five children. L.L. SR., the husband, showed a definitely increased number of target cells and, correlated with this, a decreased fragility. His MCV and MCH were somewhat low. It is probable that he represents a mild case of thalassemia minor, but the findings are certainly borderline. The fact that this individual was from out of town made repetition of the studies impossible. His son, L.L., JR., likewise presents a borderline picture. We feel that it is impossible to classify him definitely, but that he is probably normal. The problem presented by these borderline cases will be returned to later.

(11) *The L. kinship.* The proband, J.L., #240313, was a 28 year old married male admitted to the hospital because of infectious hepatitis. The blood findings were typical of thalassemia minor, and the target cell picture was especially striking. However, this patient represented an interesting diagnostic problem, because infectious hepatitis is also characterized by the presence of target cells (GREENBLATT, 1943), and it is not known how much more of the thalassemia minor picture in addition to the occurrence of target cells is duplicated. It seems unlikely that an acute infectious process could alter the cell

constants to the extent seen here, i.e., while the MCV might temporarily diminish (cf. VALENTINE and NEEL, 1945), it is improbable that hemoglobin would be extruded and a low MCH result. Moreover, the patient's mother showed findings typical of thalassemia minor. It is therefore felt that this patient represents the occurrence of infectious hepatitis in an individual with thalassemia minor. The findings on the patient, his mother, step-father, step-sister, maternal grandparents, and an uncle are given in table 11 and figure 1k.

TABLE 15

Hematological findings in the SA. *kinsnip.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
J.S., proband	48	15.0	6.49	45.6	70.2	23.1	32.9	3+	1+	0.44 co.44	0.22 0.32
A.S., wife	43	15.0	4.87	41.0	84.1	31.6	36.6	0	0	0.44 co.44	0.32 0.32
F.S., daughter	18	15.2	5.445	46.0	84.4	27.9	33.0	0	0	0.44 co.48	0.32 0.32
A.S., son	14	12.7	6.06	39.5	65.2	21.0	32.1	4+	1+	0.44 co.48	0.20 0.32

(12) *The C. kinship.* The proband, M.C., #174689, was a four year old girl admitted to the hospital because of acute pharyngitis and follicular tonsillitis, and bronchopneumonia, from which she made an uneventful recovery. Thalassemia minor was an incidental finding. Observations on the patient and her parents are summarized in table 12 and figure 1l.

(13) *The Sy. 2 kinship.* The proband, E.SY., was a two year old boy who entered the hospital with an acute laryngo-tracheo-bronchitis. Routine examination of the blood revealed thalassemia minor. The results of studies on the patient, his parents, and a maternal aunt are given in table 13 and figure 1m.

(14) *The La. kinship.* The proband, G.LA., a 32 year old white woman, was found to have thalassemia minor during the survey of the Y family reported on in VALENTINE and NEEL (1944); she is the G.L. of table 1 and figure 1. She had three children, one of whom, a 14 year old boy, had been followed in Pediatric Clinic for several years because of epilepsy. His hematologic findings are summarized in table 14 and figure 1n. The father of the family was unusually uncooperative and refused to allow himself or the other two children to be examined.

(15) *The Sa. kinship.* The proband, J.SA., #243556, was a 48 year old man who entered the hospital with a partial left hemiplegia. Thalassemia minor was detected during a survey of the blood smears of hospital patients of Italian extraction. The findings on the patient, his wife, and the two of their seven children who were available, are given in table 15 and figure 1o.

(16) *The Cot. kinship.* The proband, J.COT., #146775, was a seven year old

girl admitted to the hospital for the study of a persistent cervical lymphadenopathy. Thalassemia minor was detected during a survey of hospital patients. Data are presented on the parents and two siblings (table 16 and fig. 1p). The family is unusual in several respects. The father shows a mild polycythemia, with MCV and MCH on the low side. The mother (divorced), thought to be the parent from whom the proband inherited the disease, shows an exceptionally high hematocrit (45.3) for a woman with this condition. Her MCV is on the low side, but within normal limits. Her MCH is definitely de-

TABLE 16

Hematological findings in the COT. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
J.C., proband	7	11.5	5.175	36.9	71.3	22.2	31.1	3+	3+	0.42 co.50	0.22 0.34
J.C., father	37	17.3	6.585	50.5	76.7	26.5	34.2	0	0	0.48 co.46	0.28 0.30
B.P., mother	33	13.2	5.80	45.3	78.0	22.8	29.1	2+	2+	0.44 co.46	0.24 0.30
V.C., brother	12	14.5	5.115	40.5	79.2	28.3	35.8	0	1+	0.46 co.46	0.32 0.30
A.C., sister	9	14.5	5.32	43.0	80.0	27.3	33.7	0	1+	0.44 co.46	0.28 0.30

creased. The appearance of her smear plus the results of the fragility test make the diagnosis of thalassemia minor highly probable. Finally, both siblings showed more ovalocytosis than is commonly encountered.

(17) *The Po. kinship.* The proband, J.Sc., #19247, was a boy of four who died in this hospital in 1933 with thalassemia major. The blood findings in this boy's parents and siblings have been reported previously (VALENTINE and NEEL, 1944). Since then it has been possible to carry out hematological studies on 10 additional members of the family. With one exception they were all relatives on the maternal side. Study of the grandfather, J.P., was restricted to the examination of a blood smear. This was typical of thalassemia minor, but in the absence of confirmatory evidence the diagnosis is of course tentative. We have also restudied J.Sc., listed as a 6 year old girl in the previous paper, because her blood findings were equivocal at that time and further information was desired. The present findings on this girl confirm the earlier ones in all essential respects—she has slightly microcytic, hypochromic erythrocytes whose fragility is minimally (but insignificantly) decreased, and shows slight changes in her blood smear. The findings are consistent with thalassemia minor, but she could also be a "normal" overlap. It is interesting that J.Sc.'s mother—who because of the birth of a son with thalassemia major is certainly a minor—shows one of the mildest pictures yet encountered, the

only significant departure from normal being an increased number of ovalocytes. However, the mother's siblings showed the typical picture of thalassemia minor.

Analysis of the Data[3]

The most common type of sibship represented in the foregoing data is that in which the proband member of the sibship is a child with thalassemia minor,

TABLE 17

Hematological findings in the PO. *kinship.*

NAME	AGE	Hgb	RBC	HEMAT.	MCV	MCH	MCHC	TARGET CELLS	OVAL CELLS	HEMOLYSIS	
										BEGINS	COMP.
J.S., sister	9	13.1	5.66	40.2	71.0	23.1	32.6	0	1+	0.46 co.50	0.30 0.34
P.DEM., aunt (M)	50	13.5	6.25	40.6	65.0	21.6	33.2	2+	2+	0.44 co.46	0.26 0.32
L.DEM. husband of P. DEM.	56	14.8	4.62	43.2	93.4	32.0	34.2	0	0	0.46 co.48	0.32 0.32
J.DEM., cousin	22	14.2	4.54	41.2	90.8	31.3	34.5	0	0	0.46 co.48	0.32 0.32
S.C., aunt (M)	48	13.7	4.815	39.5	82.0	28.5	34.7	0	1+	0.46 co.48	0.32 0.34
F.PO., uncle (M)	45	14.3	6.625	41.9	63.2	21.6	34.1	2+	1+	0.44 co.50	0.20 0.34
C.PO., wife of F.PO.	41	13.6	4.12	39.0	94.6	33.0	34.9	1+	0	0.44 co.46	0.28 0.32
J.PO., cousin	21	17.2	5.265	46.7	88.6	32.7	36.8	0	0	0.46 co.46	0.32 0.32
P.SC., uncle (P)	50	15.3	6.80	46.0	67.6	22.5	33.3	2+	0	0.46 co.48	0.26 0.34
C.PO., uncle (M)	34	14.8	7.525	46.3	61.5	19.7	32.0	1+	3+	0.46 co.46	0.28 0.32

both of whose parents have been studied, with one known to be normal and one known to have thalassemia minor. Since sibships of this type always contain at least one affected member, no matter what the size of the sibship, the ratio observed in a collection of such sibships is distorted from that expected on the basis of any given genetic hypothesis. BERNSTEIN (1929) and others, especially HOGBEN (1933), have proposed a method to make allowance for this fact. In a previous publication (VALENTINE and NEEL, 1944) we found that in such sibships out of a total of 44 segregants collected from the literature and our own data, 36 had thalassemia minor, whereas expectation, if a 1:1 ratio obtained, according to the tables given by HOGBEN (1933), was 25.2.

[3] DR. SEWALL WRIGHT and DR. PAUL DAVID have kindly given us invaluable aid in the statistical treatment of these data.

The excess of thalassemia minor over expectation, 10.8 ± 2.8, appeared significant. However, HOGBEN'S method assumes complete ascertainment in a population of the trait being studied, i.e., assumes that all affected persons in the population are on record and in any investigation will all serve as probands. This is true of thalassemia major, so that the application of this method to test adherence to a 3:1 ratio in sibships in which thalassemia major is segregating, as was done previously, is valid. However, for thalassemia minor, ascertainment more nearly approximates zero. That is to say, only a small proportion of affected persons in the population has been sampled, and the chance of a segregating sibship having more than one proband is small. In the present series of 17 kinships, none had two probands with thalassemia minor. Under these circumstances it is more appropriate to use the so-called sib method (WEINBERG, 1927; FISHER, 1934) and calculate expectation with the formula

$$r_s = \sum (n_s[1 + (s - 1)p]) \pm \sqrt{[\sum n_s(s - 1)]pq}$$

where

s = number of members in sibship
r_s = expected number of affected individuals
n_s = number of s-membered sibships
p = expected proportion affected = $\frac{1}{2}$, and
$q = 1 - p = \frac{1}{2}$.

Recalculation of the previous data, using this formula, leads to an expectation on the basis of a 1:1 ratio of 28.5 affected persons, the excess, 7.5 ± 2.8, still appearing significant despite the changes in technique. This apparent excess was one reason for undertaking the present study.

The 17 kinships herein described may be broken down into 29 different sibships. Although the above described type of sibship is the most common, and one for which standard methods of analysis exist, it is only one of a number of distinct types represented in these data. In order to utilize efficiently all the data now available, it is necessary to develop a clear definition of the different types of sibships, and the ratio expected in each type, when the assumption is made that we are dealing with a single factor with distinguishable effects in homozygote and heterozygote. The following classification of the different possible types of sibships and the ratio expected in each type is perhaps more elaborate than the present data warrant, but is introduced because of its applicability to the study of segregation in small human families under certain circumstances. The calculation of expectation by the formulae that follow is valid only when ascertainment of the trait in question approximates zero.

(1) *Proband a child with thalassemia minor*, segregation studied in proband's sibship.

N = number of sibs of probands (excluding thalassemia major)
$= \sum n_s(s-1)$
r = number of sibs with thalassemia minor

A. One parent found to be normal, other affected or presumed to be affected.

$$r = .500N \pm .500\sqrt{N}$$

B. Neither parent tested, no sib with thalassemia major.

$$r = .503N \pm .500\sqrt{N}$$

C. One parent found to have thalassemia minor, other untested, no sib with thalassemia major.

$$r = .506N \pm .500\sqrt{N}$$

D. Both parents found to have thalassemia minor and/or thalassemia major is in sibship.

$$r = .667N \pm .471\sqrt{N}$$

(2) *Proband a parent with thalassemia minor*, segregation studied in proband's children.

N = number of children, excluding thalassemia major

A. Other parent found to be normal.

$$r = .500N \pm .500\sqrt{N}$$

B. Other parent untested and thalassemia major not in sibship.

$$r = .506N \pm .500\sqrt{N}$$

C. Other parent with thalassemia minor and/or thalassemia major in sibship.

$$r = .667N \pm .471\sqrt{N}$$

(3) *Proband a child with thalassemia major*, segregation studied in proband's siblings. Both parents presumed to have thalassemia minor if not tested.

N = number of children excluding thalassemia major

$$r = .667N \pm .471\sqrt{N}$$

(4) *Proband an affected child of one of sibs*, segregation studied in parental sibship.

A. The sib in question is found to have thalassemia minor (or is the parent of a case of thalassemia major) and is to be treated as if the proband under the appropriate category of class (1).

B. The sib in question has not been tested but thalassemia major is in the sibship which is to be treated as in class (3).

C. The sib in question has not been tested and thalassemia major is not present in the sibship but at least one of the parents of the sibship (grandparents of the original proband) is affected and may be treated as proband as in class (2).

D. The sib in question has not been tested, neither of his parents have been tested, and thalassemia major is not present in the sibship, but there is a member of the sibship with thalassemia minor who may be treated as the proband under (1 B).

The expectation under (1 B), (1 C), and (2 B) may be calculated approximately as follows, where

Th′ = the factor responsible for thalassemia major and minor

Th = its normal allele, and

q = the frequency of Th′ in the population.

(1 B). Proband with thalassemia minor, neither parent tested.

GENOTYPE	FREQUENCY AT BIRTH	APPROXIMATE FREQUENCY AMONG PARENTS	
Th′Th′	q^2	0	
Th′Th	$2q(1-q)$	$2q/(1+q)$	$q^2 = .0004$[4]
ThTh	$(1-q)^2$	$(1-q)/(1+q)$	$q = .02$ approximately
	1	1	

PARENTS	IN WHOLE POPULATION	EXCLUDING NORMAL × NORMAL	Th′Th PER SIBSHIP	Th′Th IN SIBSHIP UNDER (1B)
Th′Th × Th′Th	$4q^2/(1+q)^2$	q	2/3	$(2/3)q = (1/6)4q$
Th′Th × ThTh	$4q(1-q)/(1+q)^2$	$1-q$	1/2	$(1/2)(1-q) = (1/6)(3-3q)$
ThTh × ThTh	$(1-q)^2/(1+q)^2$			
	1	1		$\frac{3+q}{6} = \frac{3.02}{6} = .503$

$$r = .503N \qquad SE = \sqrt{N \times .503 \times .497} = .500\sqrt{N}$$

(1 C). Proband a child with thalassemia minor, one parent Th′Th, other unknown.

PARENTS KNOWN	PARENTS UNKNOWN	FREQUENCY	Th′Th PER SIBSHIP	Th′Th IN SIBSHIPS UNDER (1C)
Th′Th	Th′Th	$2q/(1+q)$	2/3	$8q/6(1+q)$
Th′Th	ThTh	$(1-q)/(1+q)$	1/2	$(3-3q)/6(1+q)$
				$\frac{3+5q}{6(1+q)} = \frac{3.10}{6.12} = .5065$

$$= .506N \pm .500\sqrt{N}$$

(2. B). Proband a parent with thalassemia minor, other parent untested. Same as (1 C).

The above ratios are all slightly too high because no cognizance is taken of the absence of thalassemia major in the sibship which is made one of the criteria in the classification. Proper allowance for this depends on the size of the

[4] See p. 58.

sibship and would be a complicated but usually unimportant matter. Theoretically some of the probabilities may also be modified slightly by information about other relatives (e.g., presence or absence of thalassemia minor in sibs of an unknown parent) but the effect of this would usually be small.

In table 18 a the sibships reported on in this and the preceding paper have been assigned to their proper categories, and genetic expectations calculated. Sibships in which the proband is the only child have been omitted from the

TABLE 18

Segregation into thalassemia minor and normal in eight different types of sibships. Further explanation in text.

a. Own data

TYPE OF DATA	OBSERVED THALASSEMIA MINOR	EXPECTED THALASSEMIA MINOR	OBSERVED NORMAL	EXPECTED NORMAL	TOTAL
1A	15	12.00	9	12.00	24
B	1	3.52	6	3.48	7
C	3	2.02	1	1.98	4
D	—	—	—	—	—
2A	4	6.50	9	6.50	13
B	1	0.51	0	0.49	1
C	0	0.67	1	0.33	1
3	7	6.67	3	3.33	10
	31	31.89	29	28.11	60
		χ^2=0.053 n=1	P=0.80–0.90		

b. Pooled data

TYPE OF DATA	OBSERVED THALASSEMIA MINOR	EXPECTED THALASSEMIA MINOR	OBSERVED NORMAL	EXPECTED NORMAL	TOTAL
1A	27	19.50	12	19.50	39
B	5	6.54	8	6.46	13
C	6	4.05	2	3.95	8
D	1	2.00	2	1.00	3
2A	8	9.00	10	9.00	18
B	5	4.55	3	4.45	9
C	0	0.67	1	0.33	1
3	16	15.34	7	7.66	23
	69	61.65	45	52.35	114
		χ^2=1.908 n=1	P=0.10–0.20		

calculations, since they are of no statistical value. The types of sibships included in the subgroups of (4) can all be reduced, as noted, to sibships listed under classes (1), (2), and (3), and so class (4) is not included in the table.

It should be noted that the ratios as calculated do not include probands. Because of the small numbers of individuals in any one class, errors of estimate have not been calculated for each class. Instead, the expected numbers of the two types have been totaled, and χ^2 calculated for the difference between observation and expectation. Since the proband is not included in the calculated numbers of normal and expected, χ^2 can be used, whereas the methods described previously, which include the probands in the calculated ratios, do not readily allow for the combination of groups of data in this manner. Table 18 b contains a similar computation for all the available data, that is, our own plus those in the literature. There is good agreement between observation and expectation, and the data are consistent with the hypothesis that the postulated Th′ factor is a single gene responsible when heterozygous for thalassemia minor and when homozygous for thalassemia major. The previously observed tendency towards a significant excess of thalassemia minor disappears in the light of additional data.

Quite aside from chance variations, there exist several factors which might operate to obscure the true ratios. The first factor to be considered is a possible unconscious selection of families showing a high number of affected individuals, or an unconscious selection for study of the affected rather than the normal members of a sibship. Most of the individuals with thalassemia minor whom we saw were unaware that they had hematological abnormalities. On the other hand, it is possible that those individuals in a sibship who had previously been told that they were anemic would be more anxious to be examined than those without known disease, and so would introduce a bias into the results. We are aware of only one family of this type (the S. kinship, #9), and the sibship in question was not introduced into the calculations. It is not felt that this possible bias introduces a major source of error.

The second factor is misclassification, resulting from a confusion of normal individuals with those showing mild thalassemia minor, and vice versa. It should be emphasized that the hematological diagnosis rests on three largely independent observations—the cell constants, the erythrocyte fragility, and the appearance of the stained smear. These all vary in characteristic fashion. While any one of these might be misleading because of errors in technique, it would be unusual for all three to be off simultaneously. In reviewing our data, we find three individuals whose classification is debatable. These are M.D'A., Jr., (kinship #6) and J.Sc. (kinship #17), both classified as thalassemia minor, and L.L., Jr., (kinship #10), classified as normal. In the cases of the first two, the changes are so slight that they could represent the limits of normal variation. That, on the other hand, such slight changes may indicate thalassemia minor has been shown by the case of Mrs. J.Sc., commented on previously (Valentine and Neel, 1944). She is thought to have thalassemia minor because of the birth of a child with thalassemia major. However, her cell constants are low normal, her fragility is not significantly decreased, and the chief clue to the diagnosis is a mild ovalocytosis shown in figure 3 of the previous paper.

A third factor which might tend to distort ratios is the occurrence in the population of other conditions resembling thalassemia minor. Chief among

these are the nutritional anemias of childhood, and the anemia due to chronic blood loss in the adult. The Co. twins (kinship #4) when first examined had features consistent with thalassemia minor—these later disappeared and may be presumed to have been nutritional in origin. So far as we are aware, none of the adults studied, with the possible exception, already mentioned, of S.A., SR. (kinship #3), were subject to chronic blood loss.

There do exist, then, a number of possible causes for misclassification. The same sources of error must operate in the data of others. It is not felt that these factors are present to any significant extent in the present data, that is, the agreement between observation and genetic expectation is probably not due to a fortuitous combination of distorting factors.

DISCUSSION

The solutions of problems of human heredity come slowly, and only through the combined efforts of many investigators. The data presented here buttress the homozygous-heterozygous theory of the relationship of thalassemia major and minor, by tending to erase the previously observed discrepancy between observation and expectation in marriages between normal individuals and those with thalassemia minor. However, on the basis of so small a sample, the issue cannot even now be regarded as closed. The data are also compatible with an hypothesis regarding thalassemia major as due to the interaction of two independent dominants, either of which results in thalassemia minor, a modification of the theory of McINTOSH and WOOD (1942). They were led to postulate this theory because they felt the incidence of consanguinity in the parents of patients with thalassemia major was not as high as would be expected were a rare recessive factor involved. Since then, we have shown that the incidence of thalassemia major is sufficiently high that a significant excess of consanguinity in the parents would not be expected. Unfortunately, the crucial marriage to differentiate between these two theories (thalassemia major×normal) has not been observed. In view of the inherent improbability of finding two dominants with approximately equal frequencies and similar effects, the simpler theory, of a homozygous-heterozygous relationship, seems much more probable.

One of the objectives of human heredity is to define the various subdivisions of man in terms of gene frequencies, and to arrive at some understanding of how these similarities or differences originated. Fragmentary though our knowledge of thalassemia be, enough information has been accumulated to pose some interesting questions relating to population dynamics.

To begin with, here is a situation where homozygote, heterozygote, and normal can be distinguished with a high degree of accuracy. One can, then, by direct survey establish the frequency of the genotypes ThTh, ThTh′, and Th′Th′, where Th′ represents the factor responsible for thalassemia, and Th its normal allele. In the Italian element of Rochester, N.Y., which is very predominantly of Sicilian origin, the frequency of thalassemia major has been estimated from a direct survey to be 1 in each 2,368 births (0.00042) (NEEL

and VALENTINE, 1945). The 95% fiducial limits of this estimate were found to lie at 0.00021 and 0.00076. No comparable survey of the frequency of thalassemia minor exists. DAMESHEK (1943) examined 68 consecutive blood smears on Italians in the Boston area, and found 13 (0.191) compatible with thalassemia minor. We have examined 100 consecutive blood smears of patients with Italian antecedents in Rochester, and found six (0.06) compatible with thalassemia minor. Four of these smears were followed up with complete blood and family studies—in three cases the individuals proved to have the disease. During the course of our studies we have examined 19 spouses of affected individuals—one of these (0.053) was found to have the condition. As has been pointed out previously, the frequency of thalassemia minor does not have to be determined by direct survey, but may readily be estimated once the frequency of thalassemia major is known. Such an approach results in an estimate of an incidence of 0.04, with the 5% levels of significance for the estimate lying at 0.029 and 0.056. Using this latter estimate, the frequency, q, of Th is approximately 0.98, and of Th′, 0.02.

Secondly, we possess some information concerning the selective disadvantage under which individuals with thalassemia operate. Persons with thalassemia major only very rarely reach reproductive maturity. Their selective disadvantage(s) therefore approximates 1. The selective disadvantage of individuals with thalassemia minor is more difficult to estimate. If they reach maturity the degree of their handicap from the standpoint of population dynamics is measured in mean family size, when this is compared with the mean family size of unaffected individuals. Such figures do not exist at present. Adequate clinical studies of thalassemia minor are still lacking, and such "clinical impressions" as do exist, including our own, are of little value. However, certain considerations make it probable that these individuals are handicapped in the competition with normal people. 1) Iron deficiency results in a microcytic, hypochromic anemia in many respects comparable to thalassemia minor. The effects of this anemia on general health have been studied by many investigators, and especially by MACKAY (1931), who demonstrated in infants with an iron deficiency anemia a "remarkable reduction" in the morbidity of respiratory and gastro-intestinal tract disease when iron was given in adequate amounts. By the large, however, the iron deficiency anemias which come to attention are more profound than the anemia of thalassemia minor. 2) In a population subject to periodic bloodshed, through war, accident, and child-birth, those individuals with an initially low hemoglobin are more apt to feel the effects of hemorrhage than those with normal values. In the 70 individuals with thalassemia minor whom we have now studied, the mean reduction in hemoglobin from normal values, making due allowance for age, was 2.0 ± 0.16 gms., where the standard of normal for an adult male was taken as 16 grams percent, and for an adult female, 14 grams percent (WINTROBE 1942). By way of a control, the mean departure of the 44 normal individuals seen during these studies from the accepted standards was $+0.25 \pm 0.15$ gms. It is conceivable that such an initial difference could be a significant factor

in shock from hemorrhage. 3) Finally, perhaps the best evidence that the reduced hemoglobin and hematocrit in this condition are deleterious is the fact that the body attempts to compensate, by an over-production of erythrocytes. Taking all these factors into consideration, we may conclude that an individual with thalassemia minor is probably slightly handicapped, with a selective disadvantage that may be estimated for purposes of calculation at between 0.1 and 0.0, and probably closer to the latter value.

We are now in a position to reach a first approximation to the mutation rate which must exist to maintain the present gene frequencies if the population is in equilibrium. WRIGHT (1929) has developed the formula

$$q^2(1 - 2h) - q(2 - 3h + hu) + 1 - h - \frac{u}{s} + hu = 0$$

where, in this case, q = frequency Th = 0.98
s = selective disadvantage of homozygote = 1
h = a factor which multiplied by s gives the selective disadvantage of the heterozygote = 0.1 to 0.0
and u = mutation rate of Th to Th′.

Substitution in the above formula, varying h within the limits indicated, yields the following estimates of the mutation rate:

Assumed Value of h	Estimated Value of u
0.0	0.0004
0.01	0.0006
0.1	0.0023

Such estimates, approximate though they be, indicate an order of magnitude considerably in excess of the rates predominantly encountered in Drosophila and Zea, in terms of "chromosome generations." Even if two independent dominants were involved, a mean rate of at least 0.0002 for each would be required. There exist but two other estimates of mutation rates in man. HALDANE (1935) estimated that "the rate of mutation at which the gene for hemophilia appears in the population of London . . . is about once in 50,000 human life cycles." GUNTHER and PENROSE (1935) estimated that mutation to the dominant factor responsible for epiloia occurred at a rate of between 1 in 60,000 and 1 in 120,000 in man. Both these estimates were high, and have led to the speculation that the mutation rate in man, in terms of "chromosome generations" is higher than in most other forms studied thus far. The present estimate, of at least 1 in 2500 human life cycles, gives support to this view, and the factor involved falls into the category of a relatively frequently mutating gene which, oddly enough, appears to achieve this mutations rate only in the Mediterranean stock. Only some 20 cases of thalassemia major have been reported in non-Mediterraneans, while no studies at all are available on the incidence of thalassemia minor. A similar situation may exist with respect to sickle cell anemia, where in a disease con-

fined almost entirely to negroes an equally high mutation rate must be postulated. Of the various possible theoretical explanations, that of a specific, mutability-stimulating factor, such as RHOADES (1941) has described in maize, seems most plausible. Such a postulation "solves" one problem only to create another.

Before accepting this estimate of mutation rate, it is well to inquire briefly into the effect on these calculations of another variable, the size of the effective breeding population. WRIGHT (1931) has furnished mathematical proof that in small breeding populations the effects of selection may be in part negated through random variations in gene frequencies. From the fact that thalassemia major occurs with a relatively high frequency in the Greeks, we may adduce that the disease has been present in Sicily at least since the period between 700 B.C. and the Punic Wars of the third century B.C., when Sicily was economically and culturally closely linked to Ancient Greece, and there was free exchange between the two countries. That is to say, the characteristic has probably been in the population of Sicily for at least 100 generations. During this period, with the population broken up into small breeding units, by chance in some a relatively high frequency of thalassemia could arise. However, the present Italian population of Rochester is drawn from all parts of Sicily, i.e., from many such units, and one would expect that the whims of chance would be more or less nullified when such a cross section is considered.

There exist at least three alternatives to postulating so high a mutation rate. 1) So far it has been assumed that the population is in equilibrium. The validity of this assumption may be questioned. However, if the mutation rate is much lower, and the characteristic tending to disappear, then the difficult problem of accounting for a higher frequency in times past arises. 2) The possibility of an excess of thalassemia minor in marriages involving thalassemia minor and normal has been repeatedly commented on. As the data now stand, the excess is well below the level of significance. If on the basis of a much larger sample this could be shown to be a real phenomenon, for reasons not yet clear, here is a solution to the dilemma. It is because of this fact that the point has been labored so. In the light of available data, this excess would seem of necessity related to some factor operative at the maturation of the gametes, since it is difficult to conceive of selection for the heterozygote. 3) Finally, an increased fertility of individuals with thalassemia minor, by comparison with normal individuals, would result in a lower estimate of the mutation rate. There are no data on this point. In the absence of adverse selection against thalassemia minor prior to the age of reproduction, an increased fertility of some 2% in individuals with thalassemia minor would maintain the equilibrium even if no mutation whatsoever of Th to Th′ were occurring. In the presence of any mutation—and there almost certainly is some—the increase in fertility which it is necessary to postulate would be less. The demonstration of so small an increase in fertility would require an extensive body of data, such as does not exist at the present time.

SUMMARY

Thalassemia minor is a mild, hypochromic, microcytic anemia observed in this country chiefly in families of Italian and Greek extraction. Thalassemia major is a much more serious familial condition, qualitatively similar to thalassemia minor, but quantitatively more severe, and usually terminating fatally in childhood. The two anemias are frequently associated in the same kinship, and whenever an individual with thalassemia major is found, both parents have thalassemia minor. The theory has previously been advanced that thalassemia major and minor are related as homozygote and heterozygote. In previous studies and compilations of the literature, there was observed an apparent excess of thalassemia minor over the expected 1:1 ratio in marriages between normal persons and those with thalassemia minor. The present paper presents data on 17 additional kinships. In a total of 60 segregants derived from several different types of marriages (data of this and a preceding paper; probands excluded), 31 persons had thalassemia minor, whereas expectation was 31.89. A summary of all the available data yields 114 segregants, of whom 69 were affected, whereas expectation was 61.65. χ^2 for the difference is 1.908, with $P=0.10-0.20$. Thus, in the light of further work the apparent excess of affected persons has largely tended to disappear.

The frequency of thalassemia major has previously been determined by survey methods (0.0004), and the frequency of thalassemia minor calculated from this (0.04). The selective disadvantage of thalassemia major is 1.0. The selective disadvantage of thalassemia minor may be estimated at 0.1 to 0.0. The mutation rate necessary to maintain the population at equilibrium if minor is at a selective disadvantage of 0.0 (i.e., functional recessive) is in the neighborhood of 0.0004, with higher rates required if selection operates against the heterozygotes. Such a rate would place this factor in the category of a frequently mutating gene, in terms of life cycles. Alternatives to postulating so high a mutation rate are considered.

ACKNOWLEDGEMENT

It is a pleasure to acknowledge our indebtedness to DR. J. S. LAWRENCE for substantial aid during the course of this investigation.

LITERATURE CITED

ANDRESEN, M. I., and E. R. MUGRAGE, 1936 Red blood cell values for normal men and women, Arch. Intern. Med. **58**: 136–146.

BERNSTEIN, F., 1929 Variations- und Erblichkeitsstatistik. Handb. Vererbgswiss. Lieferung 8 (I,C). 96 pp. Berlin: Bornträger.

CAMINOPETROS, J., 1938 Recherches sur l'anémie erythroblastique infantile, des peuples de la Méditerranée orientale. Ann. Méd. **43**: 27–61, 104–125.

COOLEY, T. B., and P. LEE, 1925 Series of cases of splenomegaly in children with anemia and peculiar bone changes. Trans. Amer. Pediat. Soc. **37**: 29–30.

DAMESHEK, W., 1940 "Target cell" anemia: Anerythroblastic type of Cooley's erythroblastic anemia. Amer. J. Med. Sci. **200**: 445–454.

1943 Familial Mediterranean target-oval cell syndromes. Amer. J. Med. Sci. **205**: 643–660.

FISHER, R. A., 1934 The effect of methods of ascertainment upon the estimation of frequencies. Ann. Eugen. **6**: 13–25.

FOSTER, P. C., and J. R. JOHNSON, 1931 Oxygen capacity and hemoglobin content of normal blood of men. Proc. Soc. Exp. Biol. **28**: 929–932.

GREENBLATT, I. J., and G. KAPLAN, 1943 Target cells in post-vaccinal jaundice. J. Amer. Med. Ass. **122**: 806–807.

GUNTHER, M., and L. S. PENROSE, 1935 The genetics of epiloia. J. Genet. **31**: 413–430.

HALDANE, J. B. S., 1935 The rate of spontaneous mutation of a human gene. J. Genet. **31**: 317–326.

HAMRE, C. J., and M. H. AU, 1942 Hematologic values for normal healthy men 16 to 25 years of age. J. Lab. Clin. Med. **27**: 1231–1244.

HOGBEN, L., 1933 Nature and nurture. 144 pp. New York: W. W. Norton and Co., Inc.

HUBBARD, J. P., and M. H. MCKEE, 1939 The anemia of rheumatic fever. J. Pediat. **14**: 66–73.

MACKAY, H. M. M., 1931 Nutritional anemia in infancy. Sp. Rep. Ser. Med. Res. Coun. Lond. **157**: 125 pp.

MCINTOSH, R., and C. L. WOOD, 1942 An inquiry into the genetic factor in Cooley's anemia. Amer. J. Dis. Child. **64**: 192–194.

NEEL, J. V., and W. N. VALENTINE, 1945 The frequency of thalassemia. Amer. J. Med. Sci. **209**: 568–572.

RHOADES, M. M., 1941 The genetic control of mutability in maize. Cold Spring Harbor Symposia on Quant. Biol. **9**: 138–144.

SMITH, C. H., 1942 Mediterranean (Cooley's) anemia in a youth of nineteen years observed since early childhood. J. Pediat. **20**: 370–382.

1943 Familial blood studies in cases of Mediterranean (Cooley's) anemia. Amer. J. Dis. Child. **65**: 681–701.

STRAUSS, M. B., G. A. DALAND, and H. J. FOX, 1941 Familial microcytic anemia. Amer. J. Med. Sci. **201**: 30–34.

VALENTINE, W. N., and J. V. NEEL, 1944 Hematologic and genetic study of the transmission of thalassemia. Arch. Intern. Med. **74**: 185–196.

1945 The artificial production and significance of target cells, with special reference to their occurrence in thalassemia. Amer. J. Med. Sci. **209**: 741–752.

WEINBERG, W., 1927 Mathematische Grundlagen der Probandenmethode. Z. I. A. V. **48**: 179–228.

WHIPPLE, G. H., and W. L. BRADFORD, 1936 Mediterranean disease—thalassemia (Erythroblastic anemia of Cooley). J. Pediat. **9**: 279–311.

WINTROBE, M. M., 1942 Clinical Hematology. 703 pp. Philadelphia: Lea and Febiger.

WINTROBE, M. M., E. MATHEWS, R. POLLACK, and B. M. DOBYNS, 1940 A familial hemopoietic disorder in Italian adolescents and adults. J. Amer. Med. Ass. **114**: 1530–1538.

WRIGHT, S., 1929 Fisher's theory of dominance. Amer. Nat. **63**: 274–279.

1931 Evolution in Mendelian populations. Genetics **16**: 97–159.

AN IMMUNOGENETIC ANALYSIS OF RACIAL DIFFERENCES IN DAIRY CATTLE[1]

R. D. OWEN, CLYDE STORMONT AND M. R. IRWIN
University of Wisconsin, Madison, Wis.

Received August 20, 1946

INTRODUCTION

MOST breeds of domestic animals occupy a position in the taxonomic hierarchy similar to that held by races in natural populations. The mechanisms leading to genetic differences in these two classes of populations are presumably much the same; the factor of human interference with the mating pattern and with selection in domestic populations is probably the only essential difference. An investigation of the genetic bases of breed differences is therefore an aspect of a problem of broad biological concern—the genetic nature of population differences within the limits of a species.

Genetic studies of differences between species are limited by the difficulty, in most cases the impossibility, of obtaining viable hybrids between them, and by the frequent sterility of the hybrids even if they are obtained. For the most part, the ordinary aspects of segregation and assortment are not commonly available to the investigator of genetic similarities and differences between these reproductively isolated groups, and techniques other than hybridization must be relied upon for analyses. One productive approach has been that of immunogenetics. IRWIN and CUMLEY (1943) have shown that immunological reactions could be used to characterize the cellular antigens common to two or more species of Columba and those peculiar to particular species, i.e., not shared with others. These authors have concluded that between the two broad groups of antigens, (1) those (probably numerous) common to all species of the genus considered, and (2) those (probably few) limited to a single species, there is a third class, (3) common to certain species and absent in others. This last group of antigens exhibits a complex network of relationships among the species. Since the cellular antigens, without known exception, are gene-controlled, the immunological techniques provide a basis for a genetic analysis of species similarities and differences even when hybridization is impossible. This application is based on at least two apparently rational assumptions: (1) that antigens common to two or more species are controlled by similar or identical genes, and (2) that the absence from one

[1] From the DEPARTMENTS OF GENETICS (No. 357) and VETERINARY SCIENCE, UNIVERSITY OF WISCONSIN, in cooperation with the BUREAU OF ANIMAL INDUSTRY, UNITED STATES DEPARTMENT OF AGRICULTURE. The program of which this study is a part has been aided by grants from the AMERICAN GUERNSEY CATTLE CLUB, the HOLSTEIN-FRIESIAN ASSOCIATION OF AMERICA, the ROCKEFELLER FOUNDATION, and the RESEARCH COMMITTEE of the GRADUATE SCHOOL.

species of a cellular antigen present in another indicates that the genetic factor controlling the antigen in the second species is absent or modified in the first. CUMLEY and IRWIN (1944) have correlated certain species differences of this sort with geographic range. Other studies of similar gene-controlled cellular antigens in different species have been reviewed by WIENER (1943).

The analysis of similarities and differences between breeds or races should be much less difficult than is a similar comparison of two or more species. For the most part, breeds or races within a species are entirely cross-fertile, permitting the application of standard genetic techniques in the analysis of their differences. The considerable volume of work that has been done on intraspecific differences in natural populations has been summarized by several contributors to *The New Systematics* (1940, HUXLEY, Editor), by GOLDSCHMIDT (1940), DOBZHANSKY (1941), MAYR (1942), HUXLEY (1942), and others. It has been generally concluded that differences between biologic races are the result for the most part of differences in large numbers of genes with individually inconspicuous effects, while an occasional gene with a distinctive effect shows a difference in frequency in the populations being compared. These differences in frequency range from cases in which a gene is virtually fixed in one race while an allele is fixed in another, to subtle contrasts detected only by statistical tests. Differences in genes with small effects may also involve variations in frequency of the genes concerned, rather than the fixation of distinct alleles in different populations, although inability to identify individual factors has made it difficult to produce evidence on this point.

Differences between breeds of domestic animals appear to rest on bases similar to those outlined above for races in natural populations. For example, the important size, milk-yield, and butterfat-percentage differences between Holstein-Friesian and Guernsey dairy cattle are apparently the result of differences in a large number of genes, no one of which has an effect great enough to permit its separate identification by present methods. Marked variation in these characteristics occurs within the breeds as well as between them—evidence that the factors concerned are not all fixed in either population.

In addition to the quantitative, polygenic differences there are instances of genes with recognizable effects known to differ in frequency in different breeds. In certain cases the most striking breed differences are dependent on a simple Mendelian basis; the many breeds and varieties of pigeons furnish excellent illustrative material (see LEVI, 1941). The Holstein and Guernsey dairy breeds differ most obviously in the frequencies of the gene for red and its allele, for black. The recessive red allele is fixed in Guernseys, while no red Holsteins are admitted to the breed register, the undesired red allele being present in that breed with a gene frequency between 0.07 and 0.1 (LUSH, 1945), and occurring only in the heterozygous condition in registered Holsteins.

In spite of the relative ease with which experiments designed to test the nature of inherent differences between breeds and races may be conceived, knowledge with respect to this problem is still incomplete and inadequate.

Studies on natural populations have been complicated by the prevalence of polygenic differences, with a resultant resort to purely statistical characterization of the populations, and an inability to single out to any significant extent the specific genes involved. Studies of this sort on breeds of domestic animals are practically excluded because of the expense of producing and maintaining segregating populations numerically adequate for the solution of the problems. Known factors in domestic animals of economic value have been largely those producing extreme abnormalities (cf. LERNER, 1944). These usually are rare in any breed, and have been of little value for breed comparisons. However, as in the case of species differences, the problems of intraspecific comparisons have been opened to detailed analysis by the field of immunogenetics.

BOYD (1940), in a critique of methods of classifying mankind, has concluded that among available anthropological materials only the human blood groups meet the essential requirements for an objective analysis of the course of human evolution. He cites a number of studies contributing significantly to an understanding of the "racial origins" of modern man. Antigenic differences in human serum, reported by CUMLEY and IRWIN (1943), will provide a tool for further extension of this field if the serum differences prove to be genetically determined. Cellular antigens other than those considered by BOYD, especially the **Rh** complex, also offer promise, and have been explored to some extent in this connection (for example, WIENER, *et al.*, 1945; LEVINE, 1945). Studies of species other than man have resulted in the discovery of numerous antigenic differences within the respective species, but relatively few of these have been carried to the point of identifying the differences with specific genetic factors, and these few have to date been inadequate for a consideration of population differences within species. The numerous antigens of the red blood cells of dairy cattle, with which the present discussion is to deal, constitute an exception.

FERGUSON (1941) presented limited data on the distribution of seven different antigens in several herds and breeds of cattle, emphasizing herd differences in antigen frequency within breeds, and suggesting a possibly significant difference between the incidence of antigen **I** in Guernseys and in other breeds. The present authors have published (1944) a brief discussion of more extensive data on the frequencies of 30 antigens in two breeds of cattle. The size of the sample has now approximately doubled, and a more general consideration of the situation appears to be in order.

MATERIALS AND METHODS

Thirty inherited cellular antigens of cattle have been described in previous publications (FERGUSON, 1941; FERGUSON, *et al.*, 1942). A number of additional antigens have since been discovered, but evidence of their existence has not yet been published and they will not be included in the present analysis. The technique of detecting cellular antigens in cattle has been described in detail in the papers cited above, and an example has been presented in illustrated form by STORMONT and CUMLEY (1943). Briefly, it involves the

production in cows or rabbits of immune sera against the blood cells of particular animals, and the analysis and fractionation of these immune sera by antibody absorption. In this manner, presumably unit reagents, that is, reagents containing antibodies against a single antigen, are prepared, and the bloods of a large number of animals are tested for the presence or absence of each of the corresponding antigens. Each test involves mixing two drops of a previously determined optimum dilution of the specific reagent, one drop of a 3% solution of the washed erythrocytes of the animal being tested, and one drop of fresh or fresh-frozen normal rabbit serum as complement. Occasionally, rabbits are encountered whose sera contain detectable antibodies against cattle cells. They cannot of course be used as a source of complement in the test. The results of the test are read as degrees of hemolysis, ranging from 0 (no hemolysis—the cells do not contain the antigen) to 4 (complete lysis). Most of the reagents give a fairly uniform degree of lysis of test cells having the antigen; for example, the reagents for antigen **H** rarely give more than a partial lysis of cell suspensions having **H**, even when the reagents are used in a low dilution and a considerable excess of antibody is present; whereas the **R** reagent in the dilution used in the tests regularly gives complete lysis of cells having **R**. Interpretation of the tests, therefore, depends in part on familiarity with the reagents; a "1" reaction to the **H** reagent would be interpreted as the result of the presence of **H** antigen on the test cells, while a similar, incomplete reaction to the **R** reagent would in general be gravely suspected as due to some factor in the tests other than the presence of **R** antigen on the test cells. Further testing, by the use of other **R** reagents and by absorption of the **R** reagent with the test cells to see whether **R** antibodies are removed, would be required in such a case before it could be concluded that the test cell definitely had **R** or lacked it. A consistent effort has been made to produce reagents clearly unit in character. Occasional questionable reactions, however, continue to occur in tests with several of the reagents, and the quantity and condition of the blood to be tested are not always such as to permit rigorous checks on these reactions. These uncertain tests are omitted from the present discussion of breed differences; they are not sufficiently frequent to affect the conclusions in this study.

The 30 antigens were assigned alphabetic symbols in the order of their discovery from **A** to **Z** and then back through the alphabet from **A′** to **H′**. No relation between antigen **A** and antigen **A′**, **C** and **C′**, etc, was implied in this nomenclature, nor does any known relationship exist between the antigens of cattle and those similarly designated in man or other species. Gaps in the alphabetic list of cattle antigens (for example, there are none labelled **D** or **L**) for the most part represent duplicates or minor variants of previously discovered antigens, at first assigned a distinctive symbol and later recognized as duplicates. Thus the thirty antigens listed are immunologically distinct. They are not all, however, independent; antigen **E**, for example, has not occurred in an animal lacking **C** (FERGUSON, 1941), and **K** has been found only in animals having both **B** and **G** (STORMONT, *et al.*, 1945). These

associations appear to have their bases in relationships among the genetic factors controlling the associated antigens. Thus an allelic series of three genes has been postulated to explain the **C-E** relationship (FERGUSON, 1941), and a series of five alleles to govern the **B-G-K** series (STORMONT *et al.*, 1945). Closely linked genes, rather than allelic series, remain a possible alternative interpretation of these associations.

There are numerous additional genetic relationships among the 30 antigens, not evidenced by consistent immunological associations. To cite a single example (STORMONT *et al.*, 1945) antigens **B** and **G** can be present together or both can be absent in an animal, or either antigen can occur without the other. As suggested above, however, the factors controlling these antigens appear to be part of a complex allelic series. The listing of the several additional antigens that are also part of this **B-G-K** complex, and the evidence that a number of other antigens are members of the **C-E** series, as well as the existence of other probable allelic series involving two or more antigens, are of no direct concern in this paper.

EXPERIMENTAL DATA

The investigations of the hereditary basis of the cellular antigens involved testing the bloods of a rather large number of animals of known parentage, together with their sires and dams. While many of these were drawn from Wisconsin herds (the UNIVERSITY herds and the PABST herd at Oconomowoc, Wisconsin, for example), many samples were also received from large herds outside the state (CARNATION FARMS, Carnation, Washington, and BLAKEFORD FARMS, Queenstown, Maryland, are two of several). Extensive additional data were drawn from tests of the blood of purebred animals involved in cases of questionable parentage. Numerous animals in all sections of the United States were included in such tests. In view of the large number of animals comprising the two breed samples, and their widespread geographic distribution, these samples are considered as providing reasonably adequate characterizations of the two breeds. Data on other breeds are too limited at present to be considered representative.

The nature and significance of breed differences

Data on the incidence of the 30 antigens are presented in Table 1, summarizing the results of tests on 1,300 Guernseys and 830 Holsteins. Differences in the total number of tests for the respective antigens as shown in the table result from four factors: (1) the omission of questionable reactions referred to earlier as constituting a very minor part of the variation in total number of tests; (2) temporary exhaustion of the supply of certain of the test reagents during the period covered by this summary—for example, no **E** reagent was available for a time in 1943, and tests for **E** are therefore less numerous in both breeds than are tests for **C** and **G**; (3) inability to find or produce a duplicate source of a reagent limited in quantity, so that tests for certain antigens had to be discontinued during the course of the study—for example, note the smaller

number of tests for **M**, **N**, and **G′**; and (4) the fact that a greater number of animals was tested for the antigens identified early in the study than for those found subsequently, explaining the general decline in number of animals tested for the successive antigens.

Considering first the separate breed characterizations as demonstrated in the table, it will be noted that the different antigens show a wide range of frequencies within each breed. Antigen **A** was found in 1,124 of the 1,299 Guernseys tested for it, a percentage incidence of 86.5%. This in Guernseys

TABLE 1

Antigen Frequencies in Holstein-Friesian and Guernsey Cattle

ANTIGEN	GUERNSEY			HOLSTEIN			BREED SHOWING HIGHER* FREQUENCY OF ANTIGEN
	TOTAL TESTED	ANTIGEN PRESENT	% HAVING ANTIGEN	TOTAL TESTED	ANTIGEN PRESENT	% HAVING ANTIGEN	
A	1299	1124	86.5	825	383	46.4	G
B	1299	776	59.7	826	341	41.3	G
C	1299	768	59.1	827	442	53.4	G
E	1225	611	49.9	768	328	42.7	G
G	1292	749	58.0	828	350	42.3	G
H	1286	201	15.6	825	104	12.6	N.S.D.†
I	1300	569	43.8	830	46	5.6	G
J	1285	503	39.2	800	289	36.1	N.S.D.
K	1270	575	45.3	781	58	7.4	G
M	851	158	18.6	460	89	19.4	N.S.D.
N	854	181	21.2	450	122	27.1	H‡
O	1290	442	34.3	764	277	36.3	N.S.D.
P	1172	52	4.4	768	8	1.0	G
Q	1254	147	11.7	792	13	1.6	G
R	1173	132	11.3	725	69	9.5	N.S.D.
S	1228	824	67.1	749	153	20.4	G
T	1178	242	20.6	743	29	3.9	G
U	1140	38	3.3	706	77	10.9	H
V	1137	211	18.6	705	162	23.0	H‡
W	1128	746	66.1	705	263	37.3	G
X	1128	181	16.0	701	68	9.7	G
Y	1137	461	40.6	715	372	52.0	H
Z	1139	910	79.9	724	397	54.8	G
A′	1133	347	30.6	700	251	35.9	H‡
C′	1098	358	32.6	681	109	16.0	G
D′	1050	141	13.4	582	179	30.8	H
E′	1092	591	54.1	693	197	28.4	G
F′	928	338	36.4	455	233	51.2	H
G′	494	309	62.6	337	142	42.1	G
H′	632	278	44.0	403	315	78.2	H

* Unless otherwise noted, $p < .01$ (chi-square test—see text).

† N.S.D. = No Significant Difference.

‡ $.01 < p < .05$.

is the most frequent of the listed antigens. The remaining 29 antigens occur with different frequencies, with **U**, the least frequent antigen in Guernseys, showing a percentage incidence of 3.3%. Nine of the antigens (**A**, **B**, **C**, **G**, **S**, **W**, **Z**, **E′** and **G′** are found in more than half of the Guernseys tested, while the remaining 21 are each found in less than half. Among the Holsteins, the most frequent antigen is **H′**, with a percentage incidence of 78.2% and the least frequent is **P** (1.0%). Only five (**C**, **Y**, **Z**, **F′** and **H′**) exceed the 50% level in Holsteins; it will be noted that two of these (**C** and **Z**) are also above this level in Guernseys. Over the entire series there is an apparent tendency for those cellular components relatively frequent in one breed to be relatively common in the other breed as well, and for those rare in one breed to be infrequent in the other. A rough measure of this relationship is the correlation (0.61 ± 0.12) of the two arrays of percentage incidence. This correlation is highly significant, statistically. It indicates an overall similarity between the two breeds in the percentage incidence of particular cellular characters relative to the frequencies of others in the breed. Regarded from this point of view, an essential difference between the two populations appears to be that the frequencies of known antigens are on the whole lower in Holsteins than in Guernseys. Thus the weighted mean incidence for the entire distribution is 38.4% in Guernseys, 28.1% in Holsteins.

In evaluating the reality of differences between the two breeds in the frequencies of the separate antigens, the hypothesis to be tested is whether the two breed samples could have been drawn from a single, homogeneous population. Application of the chi-square test for homogeneity (FISHER, 1934) gave an estimate of the probability that two independent samples drawn from such a population would show differences as large as or larger than those shown by the breed samples. With regard to antigen **A**, for example, it is apparent that considerably less than once in 100 times would two samples drawn from a single population be expected to differ as greatly in the incidence of this antigen as do the Holstein and Guernsey samples. It is concluded that Holsteins and Guernseys as represented by these samples cannot be considered members of a single, homogeneous population, but that they must be regarded as distinct with regard to the frequency of antigen **A**.

The results of similar analyses of the other antigens are included in the table. The two samples can be regarded as homogeneous for the respective antigens **H**, **J**, **M**, **O** and **R**, and the two parent populations are therefore considered as showing no significant differences in the frequencies of these five cellular characters. Seventeen of the antigens (**A**, **B**, **C**, **E**, **G**, **I**, **K**, **P**, **Q**, **S**, **T**, **W**, **X**, **Z**, **C′**, **E′**, and **G′**) are more frequent in Guernseys than in Holsteins, while five (**U**, **Y**, **D′**, **F′** and **H′**) are clearly more frequent in Holsteins and three more (**N**, **V** and **A′**) are probably so, falling between the 5% and the 1% levels of significance in the statistical test.

Effects of increased sample size

Reference has already been made to an earlier note (OWEN, *et. al.*, 1944) on the frequencies of these antigens in the two breeds, based on tests of 569

Guernseys and 513 Holsteins. In this previous paper the authors commented, "It remains to be seen whether or not the samples reported here are numerically adequate to characterize the two breeds. It is possible that a further accumulation of data may lead to some modification of detail in the suggested picture of breed differences in blood type." It is pertinent therefore to consider here the effects on this analysis of increased sample size. The present discussion is based on tests of 1300 Guernseys and 830 Holsteins.

The modifications necessitated by a consideration of more adequate samples are slight. The correlation of percentage arrays in the two breeds is unchanged. The antigens found to be clearly more frequent in either breed on the basis of the first samples remained so in the second, and no additional antigens were thrown into these classes by the additional data. In the intermediate range, however, some changes are suggested. Three of the antigens (**N**, **V**, and **A'**) whose frequencies in the previous paper could not be considered significantly different, now fall between the 5% and 1% levels of significance and appear to be more frequent in Holsteins. Another character, **J**, which previously appeared to be more frequent in Guernseys, between the 5% and 1% levels of probability, is now considered not significantly different in frequency in the two breeds. The levels of statistical significance are admittedly arbitrary, and in general these effects of increased sample size appear to be consistent with statistical expectation.

DISCUSSION

The antigenic structure of bovine erythrocytes, with regard to those antigens found in some cattle and absent in others, is a complex inherited characteristic. It is known to depend upon the action of genes at a number of genetic loci, and at certain of these loci there appears to have developed during the course of evolution a considerable amount of allelic differentiation (cf. WRIGHT, 1945). Were it not for the qualitatively distinct effects of these numerous factors, the present analysis would give a quantitative picture of breed differences similar in detail to that already available for other measurable characteristics. Thus on this basis it would be concluded that Holsteins and Guernseys represent distinct populations, differing in the antigenic structure of their red blood cells in much the same way as their milk differs in butterfat percentage. As stated above (p. 70), the average incidence of cellular antigens in Guernseys is 38.4%, in Holsteins it is 28.1%. A statistic of dispersion might be computed for each population, and it could be pointed out that while there is considerable overlapping between the two populations so that similar individuals could be drawn from each, the breeds as wholes are clearly different with regard to this characteristic.

Such an analysis would of course be inadequate here, because the genetic factors contributing to these differences are separately recognizable by virtue of their distinct qualitative effects. This permits the characterization of the breeds in more specific terms. The single comparison of average antigen incidence is converted into a series of 30 separate contrasts based on the frequencies of the constituent units (antigens) making up this complex char-

acteristic. None of the units discussed in this paper is limited in occurrence to either breed; they are all shared by both populations, and only the percentage of individuals having a given antigen varies from one breed to the other. In this respect the breed differences are marked; only five of the 30 antigens are similar in frequency in the two breeds, and the remaining 25 are more frequent in one breed or the other.

This picture of the genetic nature of race differences in cattle is not unique. It is, in fact, precisely consistent with DOBZHANSKY's proposed definition of races: "populations characterized by differing frequencies of the variable genes and chromosome structures." It therefore confirms the definition and is comparable to the analysis of race differences based on frequencies of gene arrangements in *Drosophila pseudoobscura* and *D. persimilis* (DOBZHANSKY and EPLING, 1944). The confirmation appears particularly convincing in view of the wide difference between the types of material involved in the two studies. The situation in cattle is, however, somewhat complicated by the existence of "allelic series" in which individual alleles seemingly control the simultaneous production of more than one antigen. The complex specificities of many of these factors result in a number of distinct "alleles" some of which have in common among their manifold effects the control of a single antigen. Thus the frequency of an antigen in a complex of this sort is not a direct measure of the frequency of a particular "allele," but reflects the total frequencies of all the "alleles" having the elaboration of this antigen in common. STORMONT *et. al.*, (in preparation) indicate that breed differences in individual "allele" frequencies appear to be even more extreme than those evident from the present consideration of individual antigen frequencies.

Complications of this sort seem likely to be encountered in other connections as genetic knowledge advances. If, as WHITING (1943) has suggested relative to sex determination in Habrobracon, an allelic locus may in certain cases be regarded as a chromosome segment within which crossing over does not occur, but composed of numerous genes, a distinction will need to be drawn between "allele frequency" and "gene frequency" in comparing races. A similar interpretation of the complex allelic series controlling the cellular antigens of cattle would permit the calculation of the basic "gene frequencies" from the antigen frequency data discussed in the present paper, while permitting the recognition of another basis of breed comparisons in the analysis of the larger "allelic" units. Further consideration of this aspect of the problem will be postponed until the allelic complexes have been more completely analyzed.

Within each of the breeds, more or less distinct subpopulations (herds, "lines," sibships, etc.) can also be distinguished on the basis of antigen frequencies. Such differences within breeds are often as great as those between the breeds. This situation, too, has its parallel in the natural populations of *Drosophila pseudoobscura* and *D. persimilis* referred to above, and from one point of view, both in Drosophila and in cattle, "there is a multitude of racial subdivisions of different orders" within the species (DOBZHANSKY and EPLING, 1944). The breed registers recognize a discontinuity between the major races

of cattle, and except for this brief reference to minor races within the breeds, it seems unnecessary here to elaborate this further.

SUMMARY

The antigenic structure of the red blood cells of cattle is a complex inherited characteristic in which two breeds, the Holstein-Friesian and the Guernsey, have been shown to differ. Such breed differences are not of the "all-or-none" variety, but depend on contrasting frequencies of most of the units (antigens) comprising this complex characteristic. Each of the 30 antigens included in the study is present in some members of each breed, and absent in others.

This paper confirms and extends, with minor modifications, a previous report concerning the nature of breed differences in this respect.

The presence of each antigen behaves with respect to its absence as though the two conditions were controlled by allelic genes. On this basis, the breed differences appear to depend upon contrasts in simple gene frequencies. Many of the different antigens are, however, associated in genetic complexes, and this renders the calculation of simple gene frequencies in some respects misleading. Another basis of breed comparison, here tentatively identified as differences in "allele-frequencies" as distinct from those in component "gene-frequencies," is suggested.

LITERATURE CITED

BOYD, W. C., 1940 Critique of methods of classifying mankind. Amer. J. Anthropol. **27**: 33–364.

CUMLEY, R. W., and M. R. IRWIN, 1943 Individual specificity of human serum. J. Immunol. **46**: 63–70.

1944 The correlation between antigenic composition and geographic range in the old or new world of some species of Columba. Amer. Nat. **78**: 238–256.

DOBZHANSKY, TH., 1941 Genetics and the Origin of Species. Revised edition. 446+18 pp. New York: Columbia Univ. Press.

DOBZHANSKY, TH., and CARL EPLING, 1944 Contributions to the Genetics, Taxonomy, and Ecology of *Drosophila pseudoobscura* and its Relatives. Carnegie Inst. Wash. **554**: 183 pp.

FERGUSON, L. C., 1941 Heritable antigens in the erythrocytes of cattle. J. Immunol. **40**: 213–242.

FERGUSON, L. C., CLYDE STORMONT, and M. R. IRWIN, 1942 On additional antigens in the erythrocytes of cattle. J. Immunol. **44**: 147–164.

FISHER, R. A., 1934 Statistical Methods for Research Workers. Fifth ed. 319+13 pp. Edinburgh: Oliver & Boyd.

GOLDSCHMIDT, RICHARD, 1940 The Material Basis of Evolution. 436+11 pp. New Haven: Yale Univ. Press.

HUXLEY, JULIAN (Editor). 1940 The New Systematics. 583+8 pp. Oxford: Clarendon Press.

HUXLEY, JULIAN, 1942 Evolution The Modern Synthesis. 645 pp. New York: Harper.

IRWIN, M. R., and R. W. CUMLEY, 1943 Interrelationships of the cellular characters of several species of Columba. Genetics **28**: 9–28.

LERNER, I. M., 1944 Lethal and sublethal characters in farm animals. J. Hered. **35**: 219–224.

LEVI, W. M., 1941 The Pigeon. 512+32 pp. Columbia: R. L. Bryan.

LEVINE, PHILIP, 1945 The geographical distribution of genes determining individual human blood differences. Biometrics Bull. **1**: 20–21.

LUSH, JAY L., 1945 Animal Breeding Plans. 3rd ed. 443+8 pp. Ames: The Collegiate Press.

MAYR, ERNST, 1942 Systematics and the Origin of Species. 364+16 pp. New York: Columbia Univ. Press.

OWEN, R. D., C. J. STORMONT, and M. R. IRWIN, 1944 Differences in frequency of cellular antigens in two breeds of dairy cattle. J. Animal Sci. 3: 315–321.

STORMONT, CLYDE, and R. W. CUMLEY, 1943 Cellular antigens in cattle blood. J. Hered. 34: 35–41.

STORMONT, CLYDE, M. R. IRWIN, and R. D. OWEN, 1945 A probable allelic series of genes affecting cellular antigens in cattle. (Abstract) Genetics 30: 25–26.

WHITING, P. W., 1943 Multiple alleles in complementary sex-determination of Habrobracon. Genetics 28: 365–382.

WIENER, A. S., 1943 Blood Groups and Transfusion. 438+19 pp. Springfield: Charles C Thomas.

WIENER, A. S., E. B. SONN, and R. B. BELKIN, 1945 Distribution and heredity of the human blood properties, A, B, M, N, P, and Rh. J. Immunol. 50: 341–348.

WRIGHT, SEWALL, 1945 Physiological aspects of genetics. Ann. Rev. of Physiol. 7: 75–106.

TEMPERATURE EFFECTS ON X-RAY INDUCED CHROMOSOME ABERRATIONS

KARL SAX

Harvard University, Cambridge, Mass.

Received August 25, 1946

SEVERAL investigations have shown that the frequency of X-ray induced chromosome aberrations may be changed by varying the temperature at which the nuclei are irradiated. Irradiation of Tradescantia microspores at high temperatures have produced less aberrations than did the same X-ray dose at low temperatures (SAX and ENZMANN 1939; FABERGÉ 1940; CATCHESIDE, LEA, and THODAY 1946). Such results are not attributed to the temperature effect on the incidence of breakage, but to changes in the frequency of new associations of the broken chromosomes. DARLINGTON and LACOUR (1945), on the other hand, find no temperature effect on the frequency of X-ray induced chromsome aberrations and attribute the earlier described results to the effect of temperature on the time scale of nuclear development or to changes in the nucleic acid metabolism.

The utilization of X-rays in relation to dosage, the time-intensity factor, stage of nuclear development and temperature effects should provide data of value in interpreting the nature of chromosome breakage and reunion which is a normal event in meiosis. Temperature experiments have been continued in an attempt to determine more exactly the relation between temperature and X-ray effects.

Microspores of *Tradescantia paludosa* were maintained at different temperatures shortly before—for about ten minutes—and during irradiation by immersing inflorescences in a carton of water at a given temperature. Longer temperature exposures were maintained by immersing the cartons in large bowls of water which was maintained at a relatively constant temperature. After the temperature treatment and irradiation the inflorescences were kept in cartons of water by inserting the cut stems through holes punched in the covers. These were kept in the greenhouse until the time of fixation. The greenhouse temperatures varied considerably, but each series of experiments was subjected to comparable environmental conditions. The microspores were fixed at four or five days after irradiation.

Microspores were smeared on a dry slide, fixed in alcohol-acetic and stained with acetocarmine. Analysis of chromosome aberrations was based on an examination of five or six slides with the total number of division figures ranging from 1800–3000 for the lower X-ray doses to 450–600 for microspores which had received heavy dosage. Such an analysis gives aberration frequencies which, under comparable experimental conditions, rarely deviate more than ten percent.

The experiments were designed to test the frequency of X-ray induced chromosome aberrations when the microspores were subjected to different temperatures before, during and after irradiation respectively. Microspores were subjected to the different temperatures for about ten minutes before, during, and for several minutes after irradiation. Aberration frequency of the ring and dicentric chromosomes was three to four times as large for the cold series as for the microspores irradiated at 36°. The frequency at 22° was more nearly that of the 3° series although at 400 r the number of aberrations were intermediate between the 3° and 36° series. These results, shown in table 1, are in accord with previous experiments. At 3° the aberration frequency in-

TABLE 1

The effect of different temperatures during irradiation on X-ray induced aberrations. Combined data of four experiments. 14,748 chromosomes.

DOSE	% RINGS AND DICENTRICS ×2			% ROD DELETIONS	
R. UNITS	3°	±22°	36°	3°	36°
100	1.8	1.8	0.6	0.9	0.8
120	2.2	—	0.7	—	—
200	7.1	5.5	2.3	1.8	1.4
240	11.6	—	3.3	—	—
400	29.0	18.2	6.7	4.2	3.8
Ave.	10.3	8.5	2.7	2.3	2.0

creases approximately as the square of the dosage while at 36° the exponent of the dosage curve is approximately 1.5. The high temperature seems to have an effect on X-ray dosage response similar to low X-ray intensity or fractional dosage (SAX 1941).

The simple rod deletions, which presumably arise from single breaks, differ from the 2-hit aberrations in two respects. They increase in frequency in direct proportion to X-ray dosage and they show little or no response to different temperatures during irradiation. These results are in accord with those of CATCHESIDE *et al.* and support the earlier conclusions that the incidence of breaks induced by X-rays is independent of temperature.

The temperature effect on the 2-hit aberrations seems to be due to the effect on the union of broken ends of chromosomes in new associations. Since the incidence of X-ray breakage seems to be independent of temperature the effect of temperature differences should occur after the breaks are induced. Fractional dosage and dosage intensity experiments indicate that induced breaks either undergo restitution or form new associations within a relatively short time although some breaks may remain open for perhaps hours. However most exchanges occur within a few minutes of the occurrence of the breaks. If microspores were subjected to different temperatures soon after irradiation a temperature effect might be expected. Experiments were de-

signed to test the temperature effect both during and after irradiation. The results are shown in table 2. In these experiments any change of temperature after irradiation was made as rapidly as possible but it required several minutes to transfer the inflorescences to the cartons at different temperatures and for the effective temperature change of the microspores. The results of temperature differences during irradiation were consistent and in accord with previous experiments. The effects of altered temperatures after irradiation were not consistent. The experiments were repeated many times and the average frequencies of aberrations indicate little or no effect of temperature

TABLE 2

The effect of different temperatures during and after irradiation on the frequency of X-ray induced chromosomal aberrations. Combined data of 27 experiments—97,200 chromosomes. Frequency in percent of dicentric and ring chromosomes ×2. X-ray dose 320 r at 70 r/m.

TEMPERATURE AFTER IRRADIATION	TEMPERATURE DURING IRRADIATION			AVE.
	3°	±22°	36°	
3°	21.2	15.3	9.2	15.2
22°	20.3	17.0	7.7	15.0
36°	22.5	18.1	9.8	16.8
Ave.	22.0	16.8	8.9	

after irradiation. The most variable results were obtained when the microspores were irradiated at 36° and then transferred to 3°. If there is a temperature effect after irradiation it must be limited primarily to a period of several minutes after X-ray treatment. Perhaps the variability in the results of different experiments was due in part to the variation in time of transfer from one temperature to another, but the cause of some of this variation is obscure.

If the temperature effect on aberration frequency is due to changes induced in the chromosomes the temperature changes before irradiation might be responsible for changes in aberration frequency. In our previous experiments the differential temperature treatment was started about ten minutes before irradiation and terminated several minutes after exposure. DARLINGTON and LACOUR (1945) have suggested that our earlier temperature effects were due to the effect of temperature on the nuclear cycle and nucleic acid metabolism. This conclusion seems rather naive in view of the duration of the temperature treatment, the differential response of 2-hit and 1-hit aberrations, and the apparent absence of any significant change in rate of nuclear development resulting from temperature changes after irradiation. Nevertheless we subjected Tradescantia microspores to different temperatures for one hour. They were then transferred to cartons of water at room temperature for ten minutes before raying. The results are shown in table 3, There is little or no effect of the pretreatment in relation to aberration frequency, especially when com-

pared with the effects of the same temperatures during irradiation. It is possible of course that several days or weeks of pretreatment would have an effect on X-ray sensitivity, but in our experiments the relatively short duration of temperature shock before raying seems to play no part in altering the chromosomes in relation to aberration frequency produced by X-rays.

TABLE 3

Effect of temperature differences before irradiation.
Rayed at 21°, 240 r at 70 r/m—5184 chromosomes.

PRE-TREATMENT (1 HR.)	RAYED AT	% RINGS & DICENTRICS
3°	22°	13.2
36°	22°	11.1

SUMMARY

An analysis of X-ray induced chromosomal aberrations of Tradescantia microspores shows that—(1) The frequency of ring and dicentric chromosomes is increased several times by subjecting microspores to low temperatures during irradiation although temperature differences have little effect on the frequency of rod deletions. (2) Temperature changes induced several minutes after irradiation have no consistent effect on aberration frequency. (3) Low temperatures for as long as an hour before irradiation have comparatively little or no effect on aberration frequency. The effect of temperature on X-ray induced chromosomal aberration frequency appears to be limited to the period during, or very shortly after, irradiation.

LITERATURE CITED

CATCHESIDE, D. G., D. E. LEA, and J. M. THODAY, 1946 The production of chromosome structural changes in Tradescantia microspores in relation to dosage, intensity and temperature. J. Genet. **47**: 137–149.

DARLINGTON, C. D., and L. F. LACOUR, 1945 Chromosome breakage and the nucleic acid cycle J.Genet. **46**: 180–251.

FABERGÉ, A. C., 1940 An experiment on chromosome fragmentation in Tradescantia by X-rays J. Genet. **39**: 229–248.

SAX, KARL, 1941 Types and frequencies of chromosomal aberrations induced by X-rays. Cold Spring Harbor Symp. on Quant. Biol. **9**: 93–101.

SAX, KARL, and E. V. ENZMANN, 1939 The effect of temperature on X-ray induced chromosome aberrations. Proc. Nat. Acad. Sci. **25**: 397–405.

ABSTRACTS OF PAPERS PRESENTED AT THE 1946 MEETINGS OF THE GENETICS SOCIETY OF AMERICA

BOSTON, MASS., DECEMBER 27–31, 1946

LAURENCE H. SNYDER, *Secretary*
Department of Zoology and Entomology
The Ohio State University
Columbus 10, Ohio

BISHOP, CHARLES J., Harvard University, Cambridge, Mass.: ***The genetical basis of sterility in tetraploid broccoli.***—In the consideration of sterility of an inherited nature in polyploid plants, it is necessary to modify the simple positional factor hypothesis as it is applied to ordinary diploids. Many plants, for example, are self-fertile while others are self-sterile. This problem has been investigated through a determination, by means of pollen tube growth, of cross and self-compatibility or non-compatibility in the F_2 generation of an autotetraploid broccoli, in an attempt to discover how the mode of inheritance has been changed through an artificial doubling of the chromosome complement. In broccoli the "all or none" effect of the inhibiting agent in the stigma on the development of the pollen tube makes it possible to determine this compatibility in a relatively simple technique of stigma smears. Analysis of results obtained shows that with certain plants, if used as the male, the crosses are always compatible, yet quite frequently the reciprocal cross will be non-compatible. Distinct differences appear in the behavior of those that are self-sterile, in contrast to those which are self-fertile.

BLANC, RICHARD, University of Oklahoma, Norman, Oklahoma: ***An inherited tendency to duplication of wings and antennae in Drosophila melanogaster.***—A number of individuals in stock cultures of *Gl Sb ca/Dcx F* and *Bl/Cy Gl/D* flies have been found with one or two extra antennae, posterior to and in mirror-image relationship to the normal antennae, or with a growth, often definitely wing-like in character, from the mesothorax just lateral to or replacing part of the scutellum. The mesothoracic wings and the metathoracic halteres are always present and normal in appearance. Other abnormalities present, apparently associated with the tendency to duplication, include asymmetrical reduction in size of the scutellum and various abnormalities of the head, involving missing bristles and ocelli, incomplete chitinization, incomplete fusion of the two halves of the head, reduction in size of the compound eyes, and stalked eyes. The abnormalities show a low, variable penetrance in response to temperatures above 25°C. The presence of the duplication phenomenon in two stocks derived from the same original *Gl* stock with replacement in one or the other stock of both second chromosomes, one third

chromosome and most of the other third chromosome to the right of *Gl* suggests the presence of one or more genes for the duplication character closely linked to *Gl*, or a possible, hitherto overlooked, pleiotropic effect of *Gl* itself.

BRAUCH, LIANE R., T. O. SIPPEL, KATHERINE F. HAMILTON, and W. L. RUSSELL, Roscoe B. Jackson Memorial Laboratory, Bar Harbor, Maine: *Strain specificity to transplants in the anterior chamber of the eye of the mouse.*—The reputed lack of specificity in the anterior chamber of the eye was tested by a comparison of within- and between-strain transplants of mouse ovarian tissue, using whole ovaries and fragments. Success was judged by: (1) gross observation as to persistence, (2) histological examination, (3) test for fertility of grafts by retransplantation to the ovarian capsule of the strain of origin to see whether offspring could be obtained. The third part of the experiment is not yet completed, but by the first two criteria the between-strain grafts were far less successful. This held true for grafts from adults (165 ovarian fragments), newborns (137 ovaries) and 14½ day embryos (96 ovaries). Many within-strain grafts have to date persisted for over 80 days and show follicles. Between-strain grafts regressed before the 44th day and no follicles were detectable after the 14th day.

BRAUN, WERNER, University of California, Berkeley, Calif.: *Some recent results of studies on bacterial variation.*—In the bacteriological literature "microbic dissociation" has usually been treated as a unique phenomenon of bacterial variation, since, in contrast to more simple types of bacterial variation, it is distinguished by an apparently orderly succession of variant types and the occurrence of linked character variation. However, recent demonstration of the role of inherent and environmental factors in controlling the establishment of mutants in bacterial populations has substantiated that there is no valid reason to regard so-called dissociation as different from other manifestations of discontinuous bacterial variation. Apparently orderly successive changes were shown to be due to successive establishment of mutants with inherently differential growth-rates or viability. In many instances apparently linked character changes are caused by one change only, for example, loss of capsular polysaccharide, which conditions the expression of other characters dependent thereon. In other cases, where a set of characters may undergo linked as well as non-linked variation, a single mutation affecting different levels of gene-controlled processes can be held responsible. The recognition of specific factors determining specifically selective environments has aided in understanding the differential stability of bacterial populations exhibited under certain environmental conditions and represents the basis for the investigation of a new therapeutical approach. Apart from its import on problems of taxonomy, immunology and disease control, the elucidation of factors controlling the establishment of mutants in bacterial populations has produced data which bear on some general problems of evolution.

CAMERON, JAMES W., Harvard University, Cambridge, Massachusetts:

A study of the genic control of carbohydrates in maize endosperm.—The effects of combinations of the gene *su1*, an allel called "amylaceous" (*su1*^{am}), and the dominant and recessive alleles of an independent gene "dull" (*du*) on the amounts and kinds of carbohydrates in mature maize endosperm have been measured. Due to the triploid nature of this tissue, sixteen different genotypes —including four pure-breeding and twelve intermediate ones—are obtainable. Upon a relatively isogenic background obtained by backcrossing to an inbred strain, these genotypes showed a definite pattern in regard to sugars, water-soluble polysaccharides, and starch, varying from a supersugary with three doses of each of *su1* and *du* to a pseudostarchy with three each of *su1*^{am} and *Du*. The complementary action of these two higher alleles was such that one dose each of *su1*^{am} and *Du* yielded a higher starch content than three doses of either one separately. The starch fraction of each genotype was analysed for its straight and branched-chain components, that is amylose and amylopectin. Up to 64 percent amylose was present in the most sugary types, while the more starchy ones, carrying one or more doses of both *su1*^{am} and *Du*, all contained about 28 percent amylose, as does ordinary starchy (*Su1 Du*) maize. All are in contrast to waxy maize, which produces 100 percent amylopectin (SPRAGUE et al. 1943).

CARSON, HAMPTON L., and H. D. STALKER, Washington University, St. Louis, Mo.: *A seasonal study of gene arrangement frequencies and morphology in Drosophila robusta.*—In order to determine whether cyclical variation in the frequencies of gene arrangements occurs in this species, a trapping area (approximately 50×175 yards) within a large deciduous woods near Olivette, Missouri, has been selected and intensive collections are being made at regular intervals throughout the breeding season. Of the 17 gene arrangements known for this species (in chromosome arms in which more than one arrangement is known), 14 are present in this population: three in X L, three in X R, four in II L, two in II R, and two in III R. A number of these arrangements show marked differences in north-south distribution in the eastern United States. During six collecting periods of two to four weeks duration each, extending from March 5 to August 6, 9540 chromosome arms were tested; frequencies of the various arrangements in the latter periods do not differ statistically from the earlier ones or from each other. On the whole, the frequencies of all arrangements remain remarkably constant, despite considerable building up of the population in June, followed by hot, dry July weather with an apparently severe reduction in population size. Parallel studies of the morphology of the F_2 generations from the same wild flies are being carried out; these are designed to test for the presence of morphological variation in the seasonal cycle.

CASPARI, ERNST, Wesleyan University, Middletown, Conn.: *Physiological action of the gene a in Ephestia.*—The gene *a* in Ephestia causes a reduction or inhibition of the formation of kynurenin and consequently of the pigments depending on it. Kynurenin, in the a^+ metabolism, is formed from tryptophane.

In *aa*, non-oxidized tryptophane is stored in the proteins. Since the relative amount of proteins in *aa* is not increased, the qualitative makeup of the proteins in *aa* must be different from that in a^+a^+. The enzymes oxidizing tryptophane have been studied in a^+a^+ and *aa* by measuring the O_2 consumption of homogenized tissues with and without the addition of tryptophane. It turned out that tissues of both strains are able to oxidize tryptophane at the same rate. The appearance in experiments with both strains of a dark precipitate showing the solubility behavior of skotommin, the pigment formed from kynurenin, suggests that the first reaction product in both cases is kynurenin. It appears, therefore, that in *aa* animals both the substrate and the enzyme necessary for its oxidation are present. The lack of kynurenin in *aa* animals seems to be due to an inability of substrate and enzyme to react with each other.

CHILD, GEORGE P., K. RAWLS, and W. K. HALL, University of Georgia School of Medicine, Augusta, Georgia: *Alcaptonuria through seven generations.* —Due to an inborn error of amino acid metabolism alcaptonurics are unable to to degrade completely tyrosine and phenylalanine. These amino acids are carried to the stage of homogentisic acid which the alcaptonuric excretes in the urine. The urine upon standing and becoming alkaline gradually blackens as the homogentisic acid is oxidized to a melanin-like pigment.

This defect in metabolism is usually inherited as a recessive and is alleged to be more prevalent in males. Although the disease has been known for a number of centuries, less than 200 cases have been described. The present study adds 16 hitherto unreported cases, 11 of which are still alive and whose metabolism is being studied at this laboratory. Of the 16, 10 were females.

The geneology of these alcaptonurics will be described and the relation between alcaptonuria and a number of other characters will be discussed.

CLOUDMAN, A. M., G. D. SNELL, and ELIZABETH FAILOR, Roscoe B. Jackson Memorial Laboratory, Bar Harbor, Maine: *Variations in response of nine inbred strains of mice to eight spontaneous transplantable tumors.*—The genetic theory of the transplantation of malignant tumor tissue states that the fate of implanted tumor tissue is determined by the reaction between the host and the cells of the transplanted tumor tissue. The true cancers from inbred strains will always grow readily and progressively when transplanted into other mice from the same inbred strain as the mouse that developed the original tumor. Several tumors from several different strains of mice were tested by injection into other inbred mouse strains. The following inbred strains were employed: C57 black, C57 brown (lines cd and a), C57 leaden, A albino, B alb C, C_3H, dba and P. Spontaneous tumors from four of these strains have been tested in the above lines of mice. Three of these originated in C57 black mice. They were EO771 a mammary gland carcinoma, L946AII a fibrosarcoma (originally of bone origin) and C1498 a myeloma. One mammary gland carcinoma, C617, was from a C57 brown female (line cd). Two tumors were from

dba mice. One was a mammary gland carcinoma dbrB and the other was a lymphoma P1534. There were two other tumors from the A albinos. These were a mammary gland carcinoma and a neurogenic sarcoma. The different tumors were found to vary in their ability to grow in different pure stocks of mice. Also the strains of animals tested varied in their ability to act as successful hosts to the different tumors.

CONGER, ALAN D., Harvard University, Cambridge, Mass.: ***Duration of prophase as an influence of chiasma frequency.***—Smear slides of pollen mother cells of 2N *Tradescantia paludosa* and *T. canaliculata*, and 4N *T. canaliculata* were used in this study. Slides were classified as :(1) "Early," majority of cells in prophase or earlier, only a few cells at MI, (2) "Metaphase," most cells at MI and (3) "plate," most cells developed beyond MI. A total of 3,750 cells were examined and it was found that the chiasma frequency (counted only from those cells at full MI) was lowest in the "Early" slides, higher in "Metaphases," and highest in the "Lates." If all six anthers of one bud begin development at the same time, then those cells which arrive at metaphase ahead of their sister cells (an "Early" slide) have had a shorter time in prophase than those cells remaining behind at MI after their sister cells have progressed beyond MI. It was demonstrated by smearing anthers separately that although all the cells of a single anther were in almost the same stage, other anthers of the same bud might be considerably advanced or retarded. The conclusion is made that this is a case wherein not merely a single pair of chromosomes, but numbers of entire PMC are allowed to remain in meiotic prophase a longer or shorter time than normal; that when prophase is longer more complete pairing is possible and a higher chiasma frequency of the entire nucleus results, or, the shorter the prophase the lower the chiasma frequency.

CROUSE, HELEN V., University of Pa., Philadelphia, Pa.: ***The induction of aberrant chromosome elimination in Sciara males.***—Normally Sciara sperm transmit two identical X chromosomes. Both of these are eliminated from the somatic nuclei of male embryos during cleavage and one of them is eliminated later from the germ cells. In a series of studies on reciprocal translocations it has not been possible to separate this activity of the X from its centromere: when an X translocation is taken through the sperm, it is the translocation chromosome with the X centromere that is transmitted in duplicate and later eliminated from the embryonic soma and germ line; sons are never produced by such males, presumably because of the unbalanced somatic complement that results following elimination.—Sons can now be derived from males bearing an X translocation. This is done by breeding females heterozygous for a IV-II translocation to males which transmit an X-II translocation. Approximately 5 percent of the eggs deposited by such females develop into sons whose salivary gland nuclei show a single unbroken X (maternal) and the IV-II translocation, the unbroken II as well as the two translocation chromosomes being of maternal derivation. Presumably such eggs result from 3:1 disjunc-

tion of the heterozygous translocation. The sperm of the father carrying the X-II translocation transmit two identical translocation chromosomes with the X centromere and one translocation chromosome with the centromere of II. Apparently all three of these chromosomes are eliminated from the embryonic soma. The germ cells of the sons contain an extra chromosome but so far it has not been possible to identify it, for, although the sons produce fertile sperm, none of their progeny develops beyond the first larval instar.

CUMMINGS, JEAN M., Smith College Genetics Experiment Station, Northampton, Mass, and Western Reserve University, Cleveland, Ohio: *Chromosomes of Datura ceratocaula in hybrids obtained by embryo dissection, an advance report.*—In order to obtain hybrids between the aberrant species *Datura ceratocaula* and nine other species of this genus it was necessary to dissect out the young embryos and culture them *in vitro*. When the MI of the PMC were examined in these hybrids it was found that the chromosomes formed bivalents and configurations just as they did in intraspecific crosses between races and interspecific crosses between the other more closely related species. There are 10 pairs of chromosomes and ⊙4 between *Datura discolor* and *Datura ceratocaula*. This is the closest relationship found among the chromosomes of any of the species with *Datura ceratocaula*. The hybrids between *Datura ceratocaula* and some of the other species show the following configurations; *D. ceratocaula* with *D. quercifolia* gives ⊙8+⊙4+6 bivalents; with *D. ferox* it gives a configuration of 12 and 6 bivalents; with *D. pruinosa* ⊙8+8 bivalents; with *D. leichhartii* ⊙6+9 bivalents; with *D. meteloides* ⊙8+⊙4+⊙4+4 bivalents; with *D. metel* ⊙6+⊙4+7 bivalents. There are six bivalents and a configuration of 12 in the hybrid between *Datura ceratocaula* and the standard *Datura stramonium*. The examination of the chromosomes in the hybrids between selected Prime Types of this latter species and *Datura ceratocaula* allowed the end arrangements of the chromosomes of this species to be determined.

DERMEN, HAIG, U. S. Department of Agriculture, Beltsville, Md.: *Histogenetic basis of some bud sports and variegations.*—Cytohistological studies with Datura and cranberries have shown that the shoot apex has three distinct primary histogenic layers. With colchicine, polyploidy has been induced in each layer separately. There is enough evidence to indicate that perhaps all angiosperms are characterized by distinct apical layers; dicots possibly by three layers and monocots by two. When a somatic mutation occurs, either in nature or by artificial induction, as with radiation techniques, such a mutation will be confined to one layer only. The layers remain distinct, due to the difference in the mode of cell division in the layers. In Datura it seems that cell division in L-I and L-II at the shoot apex is regularly anticlinal, and in L-III both anticlinal and periclinal. In cranberry, cell division in L-I at the shoot apex is regularly anticlinal, but in L-II periclinal divisions occur here and there. The fact that there is a great deal of independence between the apical layers in the vegetative growing point suggests that when a mutation occurs in any

layer, only a certain tisuse or tissues derived from such a layer will contain the mutation; hence, a shoot arising from a mutated area will be of the periclinal type, thus, bud sports, such as color sports of certain well-known varieties of apples, thornless mutation in blackberry, and many other types of somatic mutations in many other plants, would be periclinal in character. Gross variegation patterns observed in leaves of dicotyledonous plants, such as *Ligustrum vulgare*, *Hedera helix*, and *Euonymus japonica* var. *mediopicta*, and of monocotyledonous plants, such as *Sansevieria trifasciata* var. *laurentii*, and *Pandanus veitchii*, may be interpreted on the basis of the variability in cell division in either L-I or L-II, or both, from anticlinal to periclinal as found in the growing point of cranberry shoots. This interpretation of the cause of the gross variegational patterns must be amplified in the case of the more complex patterns, especially the mosaic ones. In such cases the presence of two plastid types occurring in various proportions among the cells is responsible for the intricate details superimposed upon the gross variegational pattern. In the monocots cell division in L-I must change from anticlinal to periclinal quite often to account for the variegation pattern characteristic of the above-mentioned plants. A similar change in L-I must account for some changes in the variegation pattern in some dicots such as *Ligustrum vulgare*.

EIGSTI, O. J., Northwestern University, Evanston, Illinois: *The pollen tube method for making comparisons of differences in mitotic rates between diploids and tetraploids*.—The technique of culture, fixation, staining, and permanent mounting of pollen tubes provides material for data relative to rates of mitosis. Details of this study are illustrated in a demonstration with slides, photographs, and charts. Each pollen tube possesses a single potential dividing cell, the generative cell, which migrates into the pollen tube and divides after a certain time following planting of pollen in the medium of culture. Since each pollen tube represents the entity of an individual, the collection of one hundred or more pollen tubes in a single colony provides a population for study and measurement. Furthermore, it is possible to collect information about differences between two populations existing under similar conditions of environment. In this case, pollen tubes of *Polygonatum pubescens* (Willdenow) Pursh, ($n = 10$), and *P. commutatum* (Schultes f.) A. Dietrich, ($n = 20$), provide different populations, and the rate of mitosis is the object for comparison. Fixations are made at 2, 3, 4, 5, and 6 hour intervals from the start of an experiment. Each pollen tube, at any interval, can be classified as a one-celled individual or a two-celled individual. Then the proportion of these classes, one-celled and two-celled, can be used as an index of the rate of mitosis. This index in turn serves as the basis for comparing the difference in rate of mitosis between diploids and tetraploids. *Polygonatum pubescens*, the diploid, shows an accelerated rate of mitosis over the tetraploid, *P. commutatum*. These observations correlate with the flowering and general growth differences existing between the species as they occur in a given locality.

FAILOR, ELIZABETH, A. M. CLOUDMAN, and G. D. SNELL, Roscoe B. Jackson

Memorial Laboratory, Bar Harbor, Maine: *Stimulation and inhibition of tumor transplants in mice by prior injections of frozen-dried tumor tissue.*—Tissue from a transplantable tumor can be reduced to a dry powder with a minimum of chemical change by drying from the frozen state. When mice are given several injections of such frozen-dried tissue, followed about 10 days later by inoculation with the living tumor, the results depend on the particular combination of tumor and strain used. Strains C57 black, C57 leaden, C57 brown, B alb C, A and dba have beer tested as hosts. Eleven tumors originating in these strains have been tested as living transplants and most of them as frozen-dried material for "immunizing" injections. Many though by no means all possible combinations have been tried. In some combinations the prior injections produce no effect on the growth of the subsequent implant, in other cases they inhibit the growth, in yet other cases they stimulate it to the point where all or nearly all mice are killed by the tumor. The results are quite specific, each tumor being most influenced by prior injections of its own tissue, though with some cross reactions, particularly where frozen-dried tissue and living tumor originated in the same strain. The results are interpreted as indicating a variety of substances (antigens?) in the frozen-dried tissue of the various transplantable tumors capable of affecting the resistance of mice to the growth of the living tumor.

FOGEL, SEYMOUR, Queens College, Flushing, New York: *Allelic differentiation and correlations in gene action.*—Anthocyanin synthesis may be induced in excised sections of green maize leaves possessing the appropriate genotype when floated on buffered glucose in the presence of light. The total amount and rate of pigment synthesized is a function, in part, of *R* action (Genetics 31: 215–216). Those alleles of *R* which effect the production of anthocyanin in the leaf collar, silks, and pericarp, etc., also catalyze a more abundant and more rapid synthesis of pigment and excised tissue cultures than those alleles which fail to initiate production of pigment in these specific plant tissues.—Experiments were designed to test the existence of any correlation of activity in the presence of added glucose with: (a) the level of action in general plant color effect; and (b) the level of action in coloration of certain specific plant tissues. Five r^r alleles of varying silk, pericarp, and general plant color effect were studied (in backcross progenies of the type R^r-Cornell/$r^r \times r^g Pl\ Pl$) with respect to the rate and total quantity of anthocyanin synthesized. The order of potency for this aspect of action does not parallel the seriation of effects found for either general plant color or for coloration of the specific tissues listed above.—In the same sense that general plant color, aleurone color, and coloration of the listed tissues represent more or less independent aspects of the action of a single genetic unit (Rec. Genet. Soc. *12*: 36–37), it is believed that the response to added glucose represents still another more or less component of action. It is not implied that these independent components of action represent independent self-reduplicating units rather than different aspects of the action of a single unit.

GAULDEN, MARY ESTHER, and J. GORDON CARLSON, National Institute of Health, Bethesda, Maryland and University of Alabama, Tuscaloosa, Alabama: *Action of different concentrations of colchicine on spindle formation and chromosome arrangement as revealed in the living cell.*—Observations on neuroblasts of the grasshopper embryo growing in culture medium containing different amounts of colchicine showed a seriation of effects as follows. (1) The lowest concentration used ($.05\times10^{-6}$ molar) has no appreciable effect on spindles, chromosomes, or mitotic rate for 5–6 hours. After this the cells react in a manner similar to that obtained immediately with the concentration of 1.9×10^{-6} molar, as described below. (2) A somewhat higher concentration (0.2×10^{-6} molar) produces spindles which are reduced in size, and often the chromosomes are not located on the cell equator. At this concentration the cells divide during the first few hours, after which they are blocked. (3) At a still higher concentration (1.9×10^{-6} molar) the spindle is extremely small, the chromosomes radiate in all directions from contiguous points of attachment on the reduced spindle, giving an appearance of clumping, and mitosis is blocked from the start of treatment. (4) At the highest concentrations used (2.5×10^{-6} through 50×10^{-6} molar) spindle formation is completely prevented, and the chromosomes are scattered at random throughout the cell. Cells in prometaphase or early metaphase at the beginning of treatment with this concentration suffer immediate destruction of the spindle, while cells in late metaphase or early anaphase exhibit division abnormalities which often lead to the formation of tetraploid cells.

GINSBURG, BENSON, and EDWARD HUTH, Roscoe B. Jackson Memorial Laboratory, Bar Harbor, Maine: *Some aspects of the physiology of gene controlled audiogenic seizures in inbred strains of mice.*—Hereditary differences in susceptibility to fatal audiogenic seizures reported by HALL were verified using inbred strains of susceptible (dba, lines 1 and 2) and non-susceptible (C57 black subline 6) mice. The symptoms suggest cholinergic involvement. Measurements of free acetylcholine reveal no interstrain differences in brains or end-organs (submaxillary glands) of normal animals. Following convulsions, the free acetylcholine content of submaxillary gland increases while that of brain remains the same. Injection of prostigmine or atropine prior to stimulation does not affect the incidence of the seizures but usually prevents their fatal termination. Eserine and acetylcholine have a similar action and may also reduce the frequency of the convulsions. Sex, or previous exposure to the auditory stimulus, has no relationship to the incidence or course of the seizures. dba mice become less susceptible with age. Animals with ruptured ear drums do not convulse upon exposure.

GINSBURG, BENSON, and FLORA KAPLAN, Roscoe B. Jackson Memorial Laboratory, Bar Harbor, Maine: *Evidence for the presence of a gene controlled oxidase in mouse skin extracts.*—0.9 percent saline extracts of mouse skins were tested for oxidative activity against a dopa substrate. Homozygous combinations of the *C* series of genes on a background for full color were used. Extracts

of *CC* and $c^{ch}c^{ch}$ animals gave activity well above the level of autoxidation but were inactive when boiled. *cc* and $c^{e}c^{e}$ extracts either showed no activity or inhibited autoxidation of the substrate. Saline extracts of several fast and slow growing melanomas originating in dba mice showed activity comparable to that obtained from dba skin. The oxidative activity of aqueous extracts of slow growing melanomas was appreciably greater than that obtained from aqueous or saline extracts of any of the skins used and higher than that of the saline extracts of the melanomas themselves.

GOODRICH, H. B., and RUTH L. HINE, Wesleyan University, Middletown, Conn.: *Interaction of genes in Lebistes reticulatus.*—The Y chromosome gene Maculatus (*Ma*) which produces a black spot on the dorsal fin has been introduced in genetic combination with the autosomal genes producing wild *BG*, blond *bG* and gold *Bg*. It has previously been shown that the phenotypic expression of wild, blond and gold involves the production of distinctive types and numbers of melanophores. The object of these experiments has been to determine how the gene, *Ma*, which in wild produces on the dorsal fin an aggregation of melanophores bordered by iridocytes, would utilize the cells provided by the autosomal genes. In combination with blond there was no aggregation of its peculiar melanophores but instead an accumulation of iridocytes and an elimination of melanophores in the same location. No effect was detected in golden. All cases were tested by backcrosses with the wild type which resulted in the reappearance of the maculatus spot in the offspring.—Tests were also made by feeding with sex hormones which showed that the presence of the female hormone (estradiol) represses the maculatus spot and the male hormone (methyltestosterone) is probably necessary for its appearance. Regeneration experiments point to the conclusion that the time of formation of the anlage of the spot is limited to a short period at the beginning of sexual maturity. Fins regenerating before this time develop the spot but it does not regenerate after maturity.

GORDON, MYRON, New York Aquarium, New York Zoological Society, New York, New York: *The three to one ratio in genetic sex determinations.*—In natural populations, the platyfish, *Platypoecilus maculatus*, has the XX=♀♀, XY=♂♂ genetic sex-determining mechanism. However, the "domesticated" stocks of the same species have the type in which WZ=♀♀, ZZ=♂♂. Since Z is probably homologous with Y, the "domesticated" type may be WY=♀♀, YY=♂♂. When a "domesticated" spotted female (*Sp*) is mated with a "wild" non-spotted male, all the daughters are spotted and all the sons are unspotted: (W)*Sp*/(Y)+ × (X)+/(Y)+=daughters, (W)*Sp*/(Y)+, (W)*Sp*/(X)+; sons, (X)+/(Y)+, (Y)+/(Y)+. By testing the spotted F1 females routinely with "wild" striped (*Sr*) males, some spotted females, group 1, (W)*Sp*/(Y)+, produced spotted daughters and unspotted sons; ratio 1♀♀.1♂♂. Some spotted females, group 2, (W)*Sp*/(X)+, when mated with the "wild" striped males (X)+/(Y)*Sr* produced three times as many

daughters as sons: (W)*Sp*/(Y)*Sr* and (W)*Sp*/(X)+ are spotted daughters; (X)+/(X)+ are unspotted daughters while (X)+/(Y)*Sr* or one-quarter are unspotted (striped) sons. Result: 3 ♀ ♀ : 1 ♂ ♂. The background of matings between "domesticated" and "wild" platyfish will be described. An explanation of the method by which the "wild" male heterogametic type of sex-determination was shifted to the female heterogametic type under domestication will be suggested.

GOWEN, JOHN W., and MARIE S. GOWEN, Iowa State College, Ames, Iowa: *Constitution of the second chromosome of Drosophila melanogaster as indicated by X-ray analysis.*—Newly produced lethal mutations in the second chromosome were examined for possible allelism, position within the chromosome, and association with translocations or inversions. The data obtained were analyzed in terms of the chromosomal size of X-ray effects and the relation these effects have to gene number in the second chromosome. Males were subjected to 2000 and 6000 r. Of 153 lethal bearing chromosomes obtained, 8 had incompleted tests, 16 carried two lethals. The 161 lethals treated had 128 occur once, 10 twice, 3 trice and 1 four times. The lethals were distributed in all chromosome regions. Close agreement between loci occupied by visible genes and the lethals was found for dumpy black, curved plexus, and plexus speck regions. Lethals were in excess in aristaless dumpy, and black purple regions. About one-third the expected lethals were found in the purple curved region. Estimates of total number of regions capable of showing lethal effects may be had from the relation $\binom{n}{142} \frac{161!}{(4!)\,(3!)^3\,(2!)^{10}} \quad \frac{1}{n^{161}}$ representing the probability of our sample. The number of these regions is 623. The variance of this estimate is 131. The fiducial limits are 426 as the lower limit and 970 as the upper limit. These results are not to be considered as estimates of gene loci but rather as average regional effects of the X-rays. The bearing of these results on gene loci in the second chromosome will be discussed.

GREEN, M. M., University of Missouri, Columbia, Mo.: *Red eye pigment in the lozenge allelic series of D. melanogaster.*—An analysis was made of the red eye pigment in adult *D. melanogaster* ♀ ♀ homozygous for the following sex-linked, recessive lozenge alleles: lz^{37}, lz^{v}, lz^{BS}, lz, lz^{34}, lz^{v4}, and lz^{3}. The brown eye pigment was removed by making each of the alleles homozygous for the gene, vermilion (*v*). Absorption spectra of extracts of the eyes of the *lz* alleles were compared with the absorption spectrum of an extract of *v* eyes. All the *lz* alleles were like *v* (and therefore wild type) in having a single red pigment absorbing maximally at 489 millimicrons. The *lz* alleles may be seriated with respect to the amount of red pigment laid down with lz^{37} having the greatest amount of red pigment and lz^{3} the least, and the other *lz* alleles falling in between. The results indicate that genes at the lozenge locus affect the quantity of red pigment laid down during development.

HAYS, F. A., Massachusetts Agricultural Experiment Station, Amherst, Mass.: *Methods of selecting breeding stock in high fecundity experiments.*—This study includes 1705 daughters of 67 sires and 222 dams, all Rhode Island Reds. Trapnest records are available on all females and the flock has been bred for high fecundity since 1916. The major objective was to find out if the first year egg record of a hen furnishes a safe criterion of her probable transmitting ability for high fecundity. The simple correlation between mothers and daughters for annual egg production was .1590 but regression was non-linear. The simple correlation between mothers' record and daughters' mean was .3064 with linear regression. Intrasire and intradam correlations were of the same magnitude, .20 and .21, suggesting that sires and dams had an equal influence on the production of daughters. These data indicate that in a flock where age at sexual maturity, absence of broodiness and high persistency are genetically stable that gross egg production is a reasonably satisfactory criterion of transmitting ability.

HOLLAENDER, ALEXANDER, and C. P. SWANSON, National Institute of Health, Bethesda, Maryland and Johns Hopkins University, Baltimore, Maryland: *Modification of the X-ray induced mutation rate in fungi by pretreatment with near infrared.*—In general the mutation rate in fungi increases linearly with increased exposure to X-rays. A significant increase of the X-ray produced mutations is found if the spores are irradiated with near infrared radiation before X-ray exposure. The infrared used covered the region from 7000 to 18000 Å with a maximum at 10000 Å. The experimental arrangement for infrared irradiation is such that the temperature rise in the culture is not more than 2 to 5° above room temperature. Infrared alone does not induce mutation. This increase of mutation rate has been observed with *Aspergillus terreus* and *Trichophyton mentagrophytes*. The significance of these findings will be discussed in relation to the increased percentage of deletions found in Tradescantia if infrared is given before or after X-ray treatment (SWANSON and HOLLAENDER, in press) and the increased percentage of chromosome rearrange- in Drosophila if infrared is given before X-ray exposure. (KAUFMANN, HOLLAENDER and GAY; *Genetics* **31**: 1946).

HOLLANDER, W. F., Larson Junior College, New Haven, Conn.: *Demonstration of pigeon genetics.*—The present status of genetics in domestic pigeons will be demonstrated by means of specimens, charts, and literature. Specimens will include plumage samples of color and pattern factors and combinations; preserved material such as lethal types; and living pigeons of several types, especially those showing behavioral or structural anomalies. Mosaic effects will be shown and their problems outlined. Charts showing linkage groups, genetics of breed origins, and auto-sexing will be presented, and a bibliography of published studies from all sources.

HULL, FRED H., Florida Agricultural Experiment Station, Gainesville,

Florida: *Theoretical regression of F_N on homozygous parents with additive or complementary gene action.*—For n uniform sets of m complementary genes let phenotypic interval of each set be $(2d)^m$, total interval $n(2d)^m$, dominance deviation of any single pair kd, bottom recessive T, and proportions of loci AA in P_1 and P_2 respectively u and w.

$$F_N = \left[\left(\frac{1}{2}+\frac{k}{2^N}\right)(P_1 - T)^{1/m} + \left(\frac{1}{2}+\frac{k}{2^N}\right)(P_2 - T)^{1/m} - \frac{k}{n^{1/m}2^N d}(P_1 - T)^{1/m}(P_2 - T)^{1/m}\right]^m + T$$

$$\frac{\partial(F_1 - T)}{\partial(P_2 - T)} = 0, \quad \text{when} \quad \frac{1+k}{2k} = \frac{(P_1 - T)^{1/m}}{n^{1/m}2d} = u.$$

When $m=1$ gene action is additive, regression of F_1 on P_1 and P_2 is curved solely by dominance. Regression of F_1 on P_2 with constant P_1 is linear and a function of P_1. If $k=1$ (dominance complete) regression of F_1 on P_2 is zero when $u=1$. If $k>1$ (overdominance or selective advantage of heterozygote), regression of F_1 on P_2 is zero when $u=(1+k)/2k$, (cf. FISHER, Genetical Theory). Regression of F_1 on P_2 is positive with lesser values of u and negative with greater values. When $m>1$ gene action is complementary (multiplicative). Regression of F_1 on P_1 and P_2 is curved by dominance and by epistasy. Regression of F_1 on P_2 with constant P_1 is curved by epistasy. The partial regression is still zero when $k=1$ and $u=1$, or when $k>1$ and $u=(1+k)/2k$. In the latter event partial regression is positive and concave downward with lesser values of u, or negative and concave upward with greater values. Partial regression is a function of both P_1 and P_2. Regression coefficients are unaffected by linkage, or by non-genetic variation.

HUTT, F. B., and R. K. COLE, Cornell University, Ithaca, New York: *The development of strains of White Leghorns genetically resistant to lymphomatosis.*—Avian lymphomatosis, apparently induced by a virus, is characterized by the accumulation of neoplastic undifferentiated lymphocytes in one or more organs, especially those of the nervous system, the viscera, or in the iris. For the past 20 years it has caused more of the prevailing high mortality among adult fowls than any other disease. Beginning with an unselected population in 1935, three strains of White Leghorns have been differentiated by selection, two of which are markedly resistant, the other extremely susceptible. Final results for 2400 females of the tenth selected generation, now under test, will be presented. Up to July 1, only about 5 percent of birds from proven sires of the resistant strains had died of lymphomatosis, whereas, in the corresponding susceptible stock, mortality from this disease was 35 percent. Diallele and reciprocal crosses for two years proved that the difference between these resistant and susceptible strains is genetic and not attributable to any cytoplasmic transmission through the egg of passive immunity in the resistant

lines, or of the causative agent in the susceptible one. The resistant strains were also bred for egg production. In tests against controls from good commercial flocks, they proved superior both in viability and egg production. Similarly, breeding cockerels of the resistant strains have demonstrated in other flocks superior ability to transmit resistance to disease and capacity for high egg production. It is considered that the results demonstrate the feasibility of controlling the disease genetically. Procedures for establishing resistance most quickly will be presented.

IRWIN, M. R., University of Wisconsin, Madison, Wisconsin: *A comparison of the antigenic characters which distinguish Pearlneck and Senegal, respectively, from Ring dove.*—The antigenic components of the red blood cells which differentiate Pearlneck (*Streptopelia chinensis*) from Ring Dove (*St. risoria*) have been obtained as units, following successive backcrosses to Ring dove of the species hybrids and selected backcross hybrids. At least nine such substances have been identified at one time or another, these being called ("d" for dove) d-1, d-2, d-3, d-4, d-5, d-6, d-7, d-8, d-9, d-10, d-11 and d-12. (A few of these substances probably are duplicates.) The genetic test for the unity of an antigenic character is that any bird possessing it should produce in backcrosses to Ring dove only two kinds of offspring: those with and those without the antigen. That is, each unit-antigen acts as if produced by one or more genes on a chromosome of Pearlneck.—By the same procedure, a minimum of ten antigenic substances which distinguish Senegal (*St. senegalensis*) from Ring dove have been identified as single entities. These are called ("s" for Senegal) s-1, s-2, s-3, s-6, s-7, s-8, s-9, s-10, s-11 and s-12. Since it has been found that Senegal has all or nearly all the antigens *common* to Pearlneck and Ring dove, and all or part of each of those which distinguish Pearlneck from Ring dove, the isolation of the antigenic characters specific to each species (Pearlneck and Senegal in contrast to Ring dove) allows a comparison of the different unit-substances of closely related species.—Thus, d-3 and s-11 appear to be homologous, as are d-4 and s-6, d-5 and s-1, and d-12 and s-9. One character, d-1, which behaves as a unit in Pearlneck, is divided into two parts (s-2 and s-12) in Senegal. One of these (s-2) is seemingly wholly contained in Pearlneck, while the other (s-12) is always combined with a substance which distinguishes Senegal from Pearlneck. The antigens d-2 and s-7 are antigenically related, as are d-11 and s-8, but not identical. In other words, a part of d-2 and s-7 are alike, but each antigen contains one or more so-called groupings which are specific for Pearlneck in one case (d-2) and for Senegal in the other (s-7). Alternative explanations of these findings and the genetic implications of the separation of these species in their evolutionary progress will be discussed.

IVES, PHILIP T., and TAYLOR HINTON, Amherst College, Amherst, Massachusetts: *Second chromosome inversions in a local wild population of Drosophila melanogaster.*—Second chromosomes derived from wild flies of the South Amherst population were analyzed for lethals by means of the standard

Bl/Cy technique and for inversions by crossing over tests with *net b cn bw* or *al b cn sp*[2]. Nine inversions were found in 131 lethal chromosomes and 13 in 139 non-lethal chromosomes. The 22 inversions were distributed 11 in the left arm and 11 in the right arm of the chromosome. The finding of 8.1 percent inversion chromosomes in this wild population is in strong contrast to the previously reported finding of no inversions occurring in one generation in 835 Canton-S laboratory stock chromosomes, suggesting either a long period of accumulation or a selective advantage of the inversion chromosomes. Cytological analysis of most of the inversions is in progress and may shed some light on this question. Comparison will also be made with inversions found in radiated chromosomes, nearly all of which are lethal when homozygous. Since it is known from previous work that about 50 percent of the wild chromosomes are lethal, there is in this case no correlation between lethals and inversions. This indicates that most of the 9 lethal chromosome inversions are probably not themselves lethal when homozygous.

KIMBALL, R. F., The Johns Hopkins University, Baltimore, Md.: *Heritable resistance to antiserum produced by antiserum and by trypsin in Paramecium aurelia.*[1]—Stock 60, variety 1 of *Paramecium aurelia* is ordinarily immobilized and killed when placed in antiserum obtained from rabbits injected with this stock. As reported by others, paramecia subjected to antiserum may develop resistance to it and transmit this resistance to descendants by fission. In my experience, resistant animals arise most frequently in serum concentrations just sufficient to immobilize nearly all the animals. Occasional animals recover spontaneously and may then be resistant to further treatment. In greater dilutions, many slightly affected animals recover but few if any prove resistant. This suggests that temporarily inherited resistance may result from nearly complete loss or inactivation of antigens by reaction with antibodies. To test this, an attempt was made to remove antigens by trypsin as LANCEFIELD has done for the M-antigen of streptococci. Following 24 hours treatment with 1 percent Difco trypsin, more than half the animals were resistant to antiserum. Controls treated with boiled trypsin were not resistant. The resistance was transmitted for fourteen fissions before finally disappearing, controls meanwhile remaining sensitive. Whatever is removed by trypsin, antigen or substance necessary for its formation, is probably formed autocatalytically; otherwise its removal should not affect the rate of its synthesis and complete restoration should occur in a very few fissions. The synthesis of antigens is also dependent on the genes since in crosses of 60 to a normally resistant stock, genic inheritance of resistance can be demonstrated.

KING, EDWARD D., Harvard University, Cambridge, Mass.: *Effects of temperature during X-radiation on Drosophila.*—Males of *D. melanogaster* were X-rayed at dosages multiples of 600 r-units from 600 to 3600 r-units at 0°C and room temperature respectively. At low temperatures for all dosages the fre-

[1] This work aided by a grant from the American Cancer Society.

quencies of induced mutations were approximately twice as great as those for the same dosages at room temperature. The viability in the cold series dropped sharply at increasing dosages.

LANDAUER, WALTER, Storrs Agricultural Experiment Station, Storrs, Conn.: *Insulin-induced phenocopies and their bearing on the causes of pleiotropism and on the problem of genetic modifiers.*—It was shown in earlier investigations that the injection of insulin into chicken eggs prior to incubation or during the early stages of development leads to the occurrence of complete or partial rumplessness. Such embryos or chicks may be considered as phenocopies of hereditary rumplessness. There are definite response differences between various breeds and stocks (White Leghorn > Light Brahma > Black Jersey Giant > Buff Orpington > "short upper beak" > Creeper). After the end of the third day of development rumplessness can no longer be produced by injection of insulin. At that time a new syndrome appears in response to treatment with insulin, namely, a shortening of the upper beak (or other beak deformities), shortening of the extremities, especially of the legs, and more rarely, hydrophthalmos. The syndrome bears close resemblance to the phenotype of the "short upper beak" lethal mutation. A homozygous "short upper beak" stock in which most of the phenotypic expression of the lethal was suppressed by selection shows high resistance toward insulin at early as well as later stages. The Creeper stock also shows little response at either period as compared with White Leghorns. The developmental mechanisms of the mutants and of insulin appear to be similar ones, the same set of modifiers protecting against both agencies. The modifiers have protective effects on systemically related parts which appear at different periods of development. The effect of insulin in late stages sheds light on the nature of pleiotropism.

LANGHAM, D. G., Instituto Nacional de Agricultura, Maracay, Venezuela: *Seedling characters in sesame* (*Sesamum indicum* L.).—An intensive study of sesame has revealed many seedling differences but only a few of them segregate sharply enough to be useful in genetical work with this plant. Those previously reported are glabrous, cupped leaves, and absence of glands on the dorsal surface of the leaves. New characters include purple, green, and yellow bud; albino, tilted cotyledons; white, purple, and green stem; and three virescents. Most of these are single gene differences, but purple bud is differentiated from green bud by complementary genes, one of which is linked with a virescent.

LANGHAM, D. G., Instituto Nacional de Agricultura, Maracay, Venezuela: *Initiation of a linkage map for sesame* (*Sesamum indicum* L.).—A genetical study of the contrasting characters in approximately 15,000 segregating populations of sesame has provided material for a preliminary mapping of more than one hundred genes. The association of the linkage groups with specific chromosomes will depend on a more critical cytological study; the haploid number of this species (*S. indicum* L.) is thirteen.

LERNER, I. MICHAEL, and L. N. HAZEL, University of California and Kimber Farms: *Population genetics of a poultry flock under artificial selection.*—Twelve years' records of the University of California poultry flock selected for egg production were analyzed. The important features of the mating system were: annual flock size from 400–700 pullets, semi-closed system of breeding, emphasis on family and progeny performance, selection and breeding from females 2 years of age or older, deliberate avoidance of close mating. The analysis indicated that selection intensity was sufficiently great to account for the observed improvement (5.6 eggs per year), if heritability of egg production is taken as 14 percent. Tested parents produced superior progeny to untested parents. The average inter-generation interval approximated two years. Ancestor elimination proceeded at a rate considerably greater than expected by chance. Thus of 77 dams of the original flock, only 5 were represented in the ancestry of the latest generation (3 out of 9 sires). Homozygosity increased by 2 percent per generation with less than expected sub-group formation. The average number of sires used per year approximated 11, while the calculated effective number was 6. The estimated standard errors of chance fluctuation of gene frequencies were 3–7 percent. The extent of inbreeding and the rôle of chance were thus greater than anticipated. There was no apparent relationship between production and inbreeding within any given year. In general, the consequences of the breeding plans used were found to be in agreement with those expected on basis of the current theories of population genetics.

LURIA, S. E., Indiana University, Bloomington, Indiana: *Non-independent mutations in bacteria.*—Mutations affecting different characters in bacteria are known to occur in most cases independently of one another. In particular, mutations affecting resistance to different bacteriophages have been shown to occur independently in *Escherichia coli* strain B (LURIA; DEMEREC and FANO). A number of complex mutations have recently been studied, however, causing resistance to several bacteriophages, resistance to each of which is produced independently by other mutations. These complex mutations may represent either different allelic changes at the same genetic sites responsible for the simple independent mutations, or changes at different genetic sites, or coupled simultaneous mutations at several genetic sites. It has also been found recently that a mutation to resistance to bacteriophage T2, which does not occur with any appreciable frequency in the wild type strain, occurs with relatively high frequency in a mutant strain already resistant to other bacteriophages. This influence of one mutation on the apparent rate of occurrence of another mutation may be an indication that the wild type allele of one gene acts as a suppressor for manifestation of the effects of the mutant allele of another gene.

MANGELSDORF, PAUL C., Harvard University, Cambridge, Mass.: *The genetic nature of teosinte.*—Annual teosinte, the closest relative of maize, with the same chromosome number as maize, differs from it primarily by a small number of chromosome segments or blocks of genes which are usually in-

herited intact and so behave like single genes. The number of these segments varies with the variety of teosinte. Nobogame teosinte differs from maize primarily by three such segments, Durango teosinte by four and Florida teosinte by four major segments and one or more smaller ones. In Florida teosinte the segments occur on chromosomes 1, 3, 4, 9 and possibly 10. Durango and Nobogame teosinte resemble Florida in having a long segment on chromosome 4. The position of the segments (in those whose position has been determined) is near the center of the chromosome and, in some cases at least, the centromere is included in the segment. In Florida teosinte two of the segments are inherited intact while two others exhibit crossing-over within the segment of 20–30 percent. All of the segments have similar genetic effects in reducing the size of seed and the number of rows of grain and in increasing the prominence of the glumes and the potential fragility of the rachis. All of the evidence so far available indicates that these varieties of teosinte are the result of hybridization of maize with another genus, probably Tripsacum but possibly a "pure" form of teosinte yet to be discovered.

MANGELSDORF, PAUL C., Harvard University, Cambridge, Mass.: *Treating genetic data with punched cards.*—The relationship between two or more genes in a test-cross is easily determined by the use of punched cards, appropriately slotted to distinguish between the dominant and recessive alleles. The method is demonstrated with data from a cross between a teosinte derivative and a maize linkage tester in which nine of the ten chromosomes are marked.

MCNUTT, C. W., University of Wisconsin, Madison, Wis.: *Expression of hereditary brachydactyly in man.*—A family having a characteristic form of brachydactyly involving shortening of the phalanges, extra segments in the phalanges, absence of phalanges (particularly in the feet), shortening of the metacarpals and modifications in the carpals, radius and ulna was studied. The condition is inherited as a dominant characterristic in two sublines and as an irregular dominant in one subline of the family. This family presents a combination of several types of brachydactyly which are inherited as distinct "types" in other families. It is suggested that the various forms of hereditary human brachydactyly are caused by the action of one dominant gene with varying expressivity and reduced penetrance. This variability in expression and normal phenotypes of some brachydactylous genotypes is thought to be due to the fact that the physical appearance of brachydactyly is a secondary result of some independent abnormality occurring in the embryo on or before the fifth week of development. Both chance and genetic milieu would thus affect the expression of this character.

MERRELL, DAVID J., Harvard University, Cambridge, Mass.: *A gene in Drosophila melanogaster affecting eye color and fertility.*—Induced by X-rays and located on the X chromosome at 0.0±, the recessive gene, called "deep orange," causes an orange eye color and in the female extremely low fertility.

Viability is rather low in both sexes but male fertility is unaffected. The females possess normal genitalia with spermathecae and tubular receptacles containing motile sperm after mating. Examination of the egg shows viable sperm to have entered the micropyle. Numerous eggs are laid, but as a result of many crosses with the wild type male a total of only 43 females and 0 males has been obtained, similar to the case reported by ANDERSON for almondex. The expected individuals are heterozygous females and males bearing the mutation on the single X chromosome. Since individuals of these genotypes are readily obtainable in expected ratios among the offspring of the cross mutant male × heterozygous female, the conclusion has been drawn that the eggs of the mutant females are deficient in some respect, but this awaits verification.

MICKEY, GEORGE H., Louisiana State University, Baton Rouge, Louisiana: *Endomitosis in tissues of the Louisiana lubber grasshopper, Romalea microptera* (Beauv.).—Increase in chromosome number, in nuclear size and in cell size occurs regularly by means of endomitosis in several somatic tissues of *Romalea microptera*. Endomitosis, as first described by GEITLER, is a modified mitotic process in which spiremes and chromosomes appear but no spindle is formed; thus sister chromatids tend to remain in bundles or groups. Since the nuclear membrane remains intact all the reduplicated chromosomes are held within the same nucleus. Consequently endomitosis results in polyploid cells. Nucleolar studies support these interpretations. At least some of these polyploid cells subsequently divide by ordinary mitosis. Testicular follicle cells, fat body cells, and probably also giant nerve cells and oenocytes undergo endomitosis.

MITTLER, SIDNEY, Illinois Institute of Technology, Chicago, Illinois: *Influence of genetic environment on the production of bristles in Dichaete Drosophila melanogaster*.—One of the effects of the Dichaete gene is a reduction in the number of bristles in the vicinity of the presutural bristle. A highly inbred Dichaete stock, with a mean bristle number (right side of the thorax) of $10.789 \pm .147$ (♂♂), was combined with various mutations. In all 18 different combinations were made and bristles counted. An attempt was made to use isogenic stocks wherever possible. The Dichaete Curly combination decreased the number of bristles to $8.970 \pm .865$ (♂♂), while Dichaete vestigial and Dichaete heldout increased the number of bristles produced respectively to $12.251 \pm .815$ (♂♂) and $12.444 \pm .148$ (♂♂). The heldout and vestigial genes decrease the general rate of development and in this manner may hinder the reduction of the thoracic bristles by the Dichaete gene. The Curly gene evidently speeds up the particular system of reactions that reduces the number of bristles. The Dichaete gene also causes the twisting of the wings so that they are held at an angle of 45°. Hence, there appears to be an interrelationship between Dichaete gene, the number of thoracic bristles and the wing mutants mentioned above.

MOSELEY, HELEN RYDQUEST, Storrs Agricultural Experiment Station, Storrs, Connecticut: *The early development of insulin-induced rumplessness in chicken embryos.*—Injection of insulin into chicken eggs before incubation produces phenocopies of genetic rumplessness (LANDAUER 1945). The embryogeny of insulin-induced rumplessness follows no specific pattern. The first indications of extreme rumplessness occur in embryos of 28 to 30 somites. The notochord bends ventrally into the proximal part of the indifferent mass of the tail bud, forming a bulge between the loci of the future allantois and caudal intestine (which is prevented from forming). The neural tube flares out and may bifurcate into dorsal and ventral branches. Later, axial elements grow into the cloaca or allantois. In less extreme cases, minor irregularities of the axial structures, particularly the notochord, become apparent at four or five days of development. It is postulated that these variations are due to (1) the individuality of the cells affected; (2) the time at which insulin acts, and the stage of differentiation of the cells; and (3) the number of cells affected. Early action on a limited number of cells might be compensated by some regulation; action at a critical developmental period would produce more drastic results; at later stages only minor aberrations might be expected. This will be shown to account for the fact that the embyrogenies of either dominant or recessive rumplessness (ZWILLING 1942, 1945) may be duplicated. It would also correlate with the findings of LANDAUER and BLISS (1946) that developmental stage at time of injection is an important factor in incidence of response.

MULLER, H. J., Indiana University, Bloomington, Indiana: *A comparison of the potentialities of individual loci for different types of visible mutations.* (Work supported by American Cancer Society grant recommended by Committee on Growth, National Research Council.)—Visible mutations, both viables and recessive lethals and steriles, affecting six selected loci—yellow, prune, white, vermillion, garnet and forked—were detected by examination of approximately 50,000 Drosophila females receiving a maternal X chromosome with these recessive markers simultaneously present and a paternal X with these genes originally normal, irradiated with 5000r in the spermatozoon stage. Striking differences occurred in incidence of observable, viable "gene-mutations." Garnet showed decidedly the most and included about $\frac{1}{3}$ subliminal except in compounds. Its peculiarities may be associated with garnet$^+$ being found cytologically by J. VALENCIA (parallel paper) and genetically by the writer to be within the region of KOSSIKOV's repeat. Excepting yellow, all loci manifested rather similar frequencies of heterozygously fertile lethals unassociated with gross rearrangement, changes interpreted as minute deletions (see VALENCIA's confirmation). Yellow, as expected, showed exceptionally few minute deletions when normal chromosomes were irradiated, doubtless because of proximity of "viability gene," but very many in sc^{L8}-sc^{8} chromosomes, because of proximity of heterochromatin, despite absence of large block. Gross rearrangement incidence varied greatly for different loci, white locus manifesting by far the most displacements to heterochromatin ("mottleds"), with

few or none to euchromatin, whereas for garnet many were to euchromatin. Thus far no significant differences have appeared in frequency of small deletions, relative to gene-mutations, resulting from radium as compared with X-rays, despite the different spacing of their "hits." This would indicate that both breaks of such deletions result from the same ultimate ion-cluster, its effects spreading over distances incompatible with the "sensitive volume" hypothesis.

NEEL, JAMES V., The School of Medicine and Dentistry and Strong Memorial Hospital, University of Rochester, Rochester, N.Y.: *An estimate of the rate of mutation of the gene responsible for thalassemia in man.*—Thalassemia minor is a mild hypochromic, microcytic anemia occurring predominantly in persons of "Mediterranean" derivation (Italians, Greeks, etc.). Thalassemia major is a similar but much more severe anemia which usually terminates fatally in childhood, and shows the same ethnological predilections. Both diseases have an hereditary basis, and from their association in kinships the hypothesis has been developed previously that thalassemia minor is due to heterozygosity for an inherited factor which when homozygous produces thalassemia major. The two anemias can be diagnosed with a high degree of accuracy. By survey, the frequency of thalassemia major in the Italian population of Rochester, N.Y., has been placed at 0.0004, from which, assuming a population in equilibrium, the frequency of thalassemia minor can be calculated approximately 0.04. The selective disadvantage of thalassemia major is 1.0. The selective disadvantage of thalassemia minor can be estimated at 0.0–0.1. From these data the minimal mutation rate necessary to maintain this population in equilibrium can be calculated to be about 0.0004, or one mutation in each 1200 life cycles. Preliminary calculations suggest a similar high rate of mutation of the gene responsible for sickle cell anemia in negroes. The occurrence of mutation rates of this magnitude in different sub-groups of man raises the possibility of "race" differences in mutation rate in man comparable to those observed in Drosophila, Zea, etc.

O'MARA, JOSEPH G., U. S. Department of Agriculture and University of Missouri, Columbia, Missouri: *The substitution of a specific Secale cereale chromosome for a specific Triticum vulgare chromosome.*—From slightly asynaptic individuals of *Triticum vulgare*, in which a bivalent from *Secale cereale* had been added to the normal 21 bivalents, individuals were secured which were monosomic for one Triticum chromosome and for the Secale chromosome. Certain of the doubly monosomic plants had speltoid characteristics which indicated that chromosome IX was the Triticum chromosome involved. Selfing these individuals produced a plant with 21II (the normal number for *T. vulgare*), which consisted of 20II from Triticum and one II from Secale. This individual was, therefore, nullisomic for chromosome IX of *T. vulgare*, and disomic for the Secale chromosome. The identity of the Triticum chromosome was verified by synthesizing the double monosome from a gamete nullisomic

for chromosome IX, combined with a gamete monosomic for chromosome IX and the Secale chromosome. This double monosome was indistinguishable from the speltoid ones secured from selfing the 22II plants described above. The effects of adding the Secale bivalent to and substituting it in the Triticum complement are quite different. Nullisomic IX plants are wholly male sterile, largely female sterile, weak, and lax. Adding the Secale bivalent to nullisomic IX, which is equivalent to substituting it for chromosome IX, restores male fertility, greatly increases female fertility, and improves plant vigor to normal. Adding the bivalent to normal *T. vulgare* results in semi-dwarf, partially female-sterile plants. These observations indicate that the effects of substitution cannot be wholly predicted from addition. More efficient specific single chromosome substitutions between genera may be possible in these and other genera.

PAULEY, SCOTT S., Harvard University, Cambridge, Mass.: *Early selection for heterosis in poplar hybrids*—A group of F1 progeny derived from the cross *Populus generosa* Henry × *P. jackii* Sargent in the spring of 1944 were selected and classed on the basis of the vigor they displayed in the early seedling stage. The plants were transplanted to the field in the spring of 1945 and grown for a second season as seedlings. In the spring of 1946 the plants were multiplied by cuttings and grown for another season in the field. Diameter measurements were made at the end of the 1944, 1945, and 1946 seasons and correlated.

Three other groups of F1 progeny were obtained by crossing *P. generosa* with *P. berolinensis* Dippel, *P. robusta* Schneider, and *P. jackii* on cut branches in the greenhouse in the spring of 1946. These plants were also classed on the basis of vigor demonstrated as young seedlings and compared with their showing at the end of the first season's growth.

Growth data is presented which indicates that selection for heterosis at such early periods is not entirely effective.

PIPKIN, SARAH BEDICHEK, Dept. of Zoology, American University of Beirut, Beirut, Lebanon: *A search for sex genes in chromosome 2 of Drosophila melanogaster.*—Sex potency of chromosome 2 was tested by the triploid method. Nine translocations involving chromosomes 2 and 4 were used; namely, 8, 45, 53, 4, 29, 40, 30, 34, and 6. The point of breakage of these translocations has been determined by the salivary gland method and published by previous authors. For end regions of chromosome 2 wild type appearing males heterozygous for a translocation and a normal 2 carrying *al sp* (or *b sp*) were crossed with y^2; *al sp* (or y^2; *b sp*) triploid females, For interior regions of chromosome 2, males heterozygous for two different translocations; the one translocation carrying *al* (or *b*); the other translocation carrying *sp*, were crossed with y^2; *al sp* (or y^2; *b sp*) triploid females. From each of these two types of crosses hyper-triploids, hypotriploids, hyper-intersexes, and hypo-intersexes could be detected in the progeny. The entire chromosome 2, divided into 10 short regions, yielded hyper-triploid females for each region, hypo-triploid females for 9

regions; hyper-intersexes for 9 regions, and hypo-intersexes for 7 regions. For 6 longer regions, hyper-triploids were obtained in all cases; hypo-triploids in 4 cases; hyper-intersexes in 4 cases; and hypo-intersexes in 3 cases. No marked influence on sexual differentiation was observed in any of the aneuploids.

RHOADES, M. M., Columbia University, New York City: *Crossover chromosomes in unreduced gametes of asynaptic maize.*—In asynaptic maize BEADLE (1930, 1933) found normal crossover values among the haploid gametes for the *sh-wx* region of chromosome 9. These haploid gametes represented a highly selected class in that they came from those sporocytes with normal metaphase pairing. On the chiasma theory of metaphase pairing these cells with 10 bivalents should have, approximately at least, the usual number of chiasmata and inasmuch as chiasmata result from crossing over they would be expected to yield normal crossover values. Diploid gametes are also produced by asynaptic plants. These presumably arise from those sporocytes with little or no association of homologous chromosomes at late prophase and metaphase. Since the failure to remain paired is due to the absence of chiasmata it might be expected that these diploid gametes would contain no or few crossover chromosomes. However tests of the genetic constitution of diploid female gametes, obtained by ascertaining the genotypes of the resulting triploid plants, revealed that crossing over had occurred in the *sh-wx* region in a considerable percentage of the tested diploid gametes. These data are not in accord with the views relating crossing over to chiasmata and chiasmata to post-diplotene association. It is possible that the diploid female gametes arise from meiocytes other than those with all or nearly all univalents but preliminary cytological studies have as yet revealed no important differences in chromosomal behavior between micro- and megasporogenesis.

RICK, CHARLES M., Division of Truck Crops, University of California, Davis, California: *A hair-suppressing gene that indirectly affects fruitfulness and the proportion of cross-pollination in the tomato.*—A single recessive gene, *dl*, affects the development of epidermal hairs on stems, petioles, peduncles, and pedicels in the following manner: the larger trichomes are reduced in size but not in number; the glandular hairs and the shorter trichomes are reduced both in size and number. The unicellular hairs that, in the normal type, bind anthers together to form the staminal tube are absent, permitting the anthers to diverge. Under field conditions *dl dl* plants yield only about ten percent as many fruits as *dl*+ and ++ plants, thus accounting for the discovery of this mutant. Artificial self-pollination restores normal fertility. The proportion of hybrid offspring indicates that nearly half of the seeds set by the *dl* type under open-pollination develop from outcrossing, whereas in the normal type outcrossing amounts to only one or two percent. Since seed production is greatly diminished, this increase in the *proportion* of hybrid offspring probably reflects a reduction in the *amount* of self-pollination to a much greater degree than an

increase in the *amount* of cross-pollination. These changes in fertility relations of the mutant are attributed to the disruption of the normal mechanism for self-pollination wrought by the separation of anthers. Certain aspects of the data pertaining to the rate of outcrossing and comparisons with fertility relations of male-sterile mutants can be explained if it is assumed that *dl dl* flowers set fruit only after pollen is transferred to them by insects.

RILEY, HERBERT PARKES, University of Kentucky, Lexington, Kentucky: *Chromosome studies in a hybrid between Gasteria and Aloe.*—In both Gasteria and Aloe the haploid complement consists of four large and three small chromosomes. Microsporogenesis was studied in an intergeneric hybrid that resembled Gasteria in its general vegetative aspect but had flowers like those of an Aloe. At first meiotic metaphase the large chromosomes showed a range of four to no bivalents. Most cells had one or two and the average was 1.3 bivalent per cell. The small chromosomes ranged from three bivalents to none with most cells having two; the average was 1.82 per cell. Therefore, there is some homology between the chromosomes of the two genera. Occasionally a univalent divides equationally at first anaphase, but usually the univalents pass undivided to one or the other pole. The distribution at anaphase agrees with that expected from metaphase configurations. A small percentage of cells at first and second anaphase had inversion bridges and fragments. No cell wall forms at first telophase. After the second division numerous "tetrads" have five or six cells. Most microspores fail to divide; the pollen grain wall forms but the protoplasm eventually disintegrates and the cell shrivels up. Less than one percent of the microspores undergo division, and the few metaphase figures show varying numbers of chromosomes. An occasional anaphase and telophase are observed, but the mature pollen appears to be 100 percent sterile.

RUSSELL, W. L., Roscoe B. Jackson Memorial Laboratory, Bar Harbor, Maine: *Splotch, a new mutation in the house mouse, Mus musculus.*—Splotch (symbol *Sp*), a new mutation in the house mouse, occurred in the C57 black strain at the Jackson Laboratory. *Sp sp* animals show white spotting on the belly and occasionally on the back. The feet are usually white and the percentage of tail spotting in the C57 black strain is increased. The gene is lethal when homozygous. *Sp Sp* embryos die in utero at approximately 14 days of age and are characterized by kinky tail, spina bifida and perhaps other abnormalities. Splotch is not an allele of *W*. It is probably linked with Fused.

SEARS, E. R., U. S. Department of Agriculture and University of Missouri, Columbia, Missouri: *The sphaerococcum gene in wheat.*—*Triticum sphaerococcum* Perc. is characterized by short culms, dense spikes, and small, spherical grains. ELLERTON showed that it differs from *T. vulgare* Vill. by a single recessive gene (or closely linked group of genes). In crosses of *T. sphaerococcum* with 17 different nullisomics of *T. vulgare* var. Chinese Spring, no F1 was of the *sphaerococcum* type, although the *sphaerococcum* gene would have been in

hemizygous condition in one of these F1's if located on any of the 17 chromosomes tested. F2 populations were grown from selected F1's, and in one F2, involving chromosome XVI of the D (or C) genome, the segregation for *sphaerococcum* type was not at random, but depended upon chromosome number: the disomic plants were *sphaerococcum*, while the monosomics resembled *vulgare*. These results are explained as follows: the *sphaerococcum* gene is located on chromosome XVI and in double dose produces the *sphaerococcum* effect; in single dose it is relatively ineffective. This unusual dosage relationship is shown also by the squarehead gene on chromosome IX, and presumably is related to the polyploid nature of the material. Chromosome XVI of *T. vulgare* increases, rather than decreases, spike and culm length, particularly between the dosages 0 and 1. Whether this effect is due to an allele of the *sphaerococcum* gene or to other factors on the chromosome is unknown.

SHIFRISS, OVED., W. Atlee Burpee Company, Doylestown, Pennsylvania: *On developmental reversal of "dominance."*—The concepts of "dominance" and "recessiveness" of traditional genetics are misleading when referred to the *static* manifestations of the heterozygote. A cross between a green-fruited vine line and yellow-fruited "bush" strain in *Cucurbita pepo* L. var. Table Queen gives in the F2 a 9:3:3:1 ratio, early in the season, and 1:3:3:9 ratio at the end of the growing season. During the period of rapid growth of the F2 population it is impossible to ascertain any genetical ratio unless characters are viewed as *dynamic* expressions. Data based on individual plant history and taken at frequent intervals during the growing season, in large F1, F2, F3 and backcross populations, indicate that the reversal in the genetical ratio is due to the reaction of the heterozygotes which, broadly speaking, manifest the characteristics of one parent early in the season and those of the other parent later on. Numerous crosses between varieties of the said species suggest that heterozygotes differ from each other in the *stage* at which the "turning point" occurs as well as in its *duration*, depending on allelic combination. Although the change, in the heterozygotes, is always from green to yellow (fruit color) and from "bush" to vine (growth habit) the reversal may not always be complete. The F2 of the cross "bush" × vine resembles superficially quantitative inheritance. This is due to the developmental reversal of dominance and to the possible greater susceptibility of the particular heterozygote to environmental modifications. The phenomenon of developmental reversal of dominance gives us a new approach to genetical analysis which has direct bearing upon the concept of genic action and heterosis.

SHULL, A. FRANKLIN, University of Michigan, Ann Arbor, Michigan: *Natural hybrids of certain subspecies of lady beetles of the genus Hippodamia.*—A hundred beetles identified as *Hippodamia quinquesignata ambigua*, typically nearly spotless, were found hibernating with a large mass of heavily spotted *H. sinuata spuria*. A few members of this group were bred and proved to be heterozygous for a spotted pattern. The probability is high that most if not

all of the others were likewise heterozygous. The source of the spotting could have been a cross with either the typical subspecies *H. q. quinquesignata*, or the subspecies *H. sinuata spuria*. Favoring the former are (1) that the genitalia of all male descendants (seven generations) had the aedeagal keel of *quinquesignata*, lacking in *sinuata spuria*, (2) that the ground color of all these descendants was red, none ochraceous buff. Favoring the latter is that they were hibernating with *H. sinuata spuria*, to which they could have been attracted by some dominant or semidominant responsiveness (perhaps to odor) which brings hibernating beetles together in single-species masses. One line of hybrids had spots four and five fused into an oblique band, which fusion is found in both *quinquesignata* and *sinuata spuria*. This band seems to exclude *H. convergens* as the source of the spotting. None of the descendants had a band across the front ends of the elytra, which might favor *sinuata spuria* as the progenitor, though this band in *quinquesignata* is chiefly determined by one gene which some spotted members of that species lack. On balance, the evidence appears to favor *H. q. quinquesignata* as the unknown parent of the hybrids.

SINGLETON, W. RALPH, Connecticut Agricultural Experiment Station, New Haven, Connecticut: *Mutations in maize inbreds.*—Mutations in maize inbreds occur more frequently than formerly supposed. Use of the pure line method in maize breeding presupposes the stability of the inbred lines used. In general this is true, but a considerable number of mutations have arisen. Purdue 39, the most widely used sweet corn inbred, has mutated to a reduced plant *rd*, (C30), which has a more cylindrical ear and an evergreen type kernel in contrast to the quicker drying P39 kernel. Also hybrids using C30 have consistently shown slightly more vigor than comparable P39 crosses. In extracted inbreds out of P39.C30 crosses several distinct variations have arisen, including defective and germless seeds, a lethal virescent seedling and a seed similar to brittle endosperm that produces lethal seedlings. All segregate in good monogenic ratios. Apparently mutations have also occurred for factors affecting combining ability. Some strains of P39 show considerable heterosis upon crossing with each other and with the reduced C30 plant. Other strains show comparatively little either among themselves or when crossed by C30.

JONES has shown that several of the standard field corn lines have mutated for morphological characters and for factors affecting combining ability associated with the morphological characters. The stable inbreds, once envisioned, are stable only for a comparatively short time and constant diligence is necessary to keep the hybrid corn yields at their high levels.

SMITH, BEN W., and MARY THOMPSON SMITH, North Carolina State College, Raleigh, North Carolina: *Sex chromosomes in Rumex hastatulus Baldw. with XY_1Y_2 pairing.*—Female plants of the dioecious species, *Rumex hastatulus* Baldw., are shown to have 8 somatic chromosomes, males, 9. In pollen mother cells at late diplotene each end of the X chromosome is paired with one of the

two Y's to form a trivalent chain. At metaphase I, the trivalent assumes a U-shape. The X regularly disjoins from the Y's and passes to the opposite pole. At metaphase II, one cell of the dyad contains 4 chromosomes (3 autosomes+X); the other contains 5 chromosomes (3 autosomes+Y_1+Y_2).

The 3 pairs of autosomes may be characterized as follows: (1) long, approximately median centromere; (2) intermediate in length, submedian centromere; (3) short, satellited, subterminal centromere. It is difficult to distinguish the X chromosomes from the longest autosomes in somatic cells. Y_1 and Y_2 are unequal in length, but resemble the second pair of autosomes. In previously investigated dioecious Rumex species, $2n=14$, female, and $2n=15$, male, are the lowest diploid numbers reported. It appears that $2n=8$ and 9 constitute a new basic number for the dioecious species of the genus.

SMITH, LUTHER, Agronomy Department, Washington State College, Pullman, Washington. *Chromosomal fragments in diploid wheat and their usefulness in genetic studies.*—Two fragments were produced (presumably) by an X-ray induced break of a chromosome at the centromere. Apparently one of the fragments subsequently reduplicated, forming a secondary trisome. Plants with the fragments are distinguishable morphologically but are fairly fertile. The fragments are rarely transmitted through the pollen and markedly affect the transmission of genes in one linkage group. In the progeny of plants with the fragments and heterozygous for a recessive gene for yellowish seedlings, 216 of 330 plants ($65\% \pm 2.6$) were yellowish. It is postulated that an interchange between one of the fragments and a normal (non-homologous) chromosome would result in associating the genes in the second linkage group with the transmission of the fragments and thus make it possible to determine with unusual ease whether a gene whose locus was unknown was in either of the linkage groups. Stocks with five such interchanges (for diploid wheat and barley, eight for maize if a corresponding tool were available), each involving one different chromosome, would make it possible to test efficiently in which of the seven chromosomes a gene was located. Such fragments (or other aberrations that affect the transmission of a chromosome) should prove useful in the study of disease resistance or other agronomic characters that are frequently difficult to analyze genetically.

SNELL, G. D., A. M. CLOUDMAN, and ELIZABETH FAILOR, Roscoe B. Jackson Memorial Laboratory, Bar Harbor, Maine: *Evidence that a large number of genes and antigens determines the compatibility of tumor transplants in mice.*—From published data on the genetics of tumor transplantation in mice it can be inferred that probably not less than 20 loci are concerned in determining the susceptibility or resistance of mice to transplantable tumors. Of several considerations that might affect this estimate, most would tend to raise rather than lower it. If, as seems probable, each of these loci acts by producing a distinct antigen, 20 or more antigens affecting tissue transplantation may be inferred. An experimental test for the existence of these antigens is possible through the

injection of frozen-dried tumor tissue prior to the inoculation of the living tumor. Injections of frozen-dried C1498 (C57 black origin) into C57 leaden mice inhibits the growth of subsequently inoculated C1498. On the other hand 15091a (A strain origin) in C57 leadens stimulates 15091a. A variety of other combinations have been tried with results varying according to the host strain, frozen-dried tumor strain, and living tumor used. The production of immunity (?) reactions with non-living tissue makes possible experiments to determine the nature of the active agents.

SONNEBORN, T. M., RUTH V. DIPPELL, and WINIFRED JACOBSON, Indiana University, Bloomington, Indiana: *Some properties of kappa (killer cytoplasmic factor) and of paramecin (killer substance) in Paramecium aurelia, variety 4.*—The gene *K* in *P. aurelia*, variety 4, works through the semi-autonomous cytoplasmic factor, kappa, to control production of a killer substance, paramecin, which kills paramecia that lack kappa. As part of a study of the action of gene *K*, the properties of kappa and paramecin are under investigation. Kappa and paramecin differ as follows: kappa is detected only in the cytoplasm, paramecin only in the fluid in which killers have lived or in sensitives exposed to this fluid; in *KK* animals, kappa is multiplied, paramecin is not; sensitives are killed by paramecin, not by kappa. When an excess of sensitives are exposed to equal volumes of a series of dilutions of paramecin, the number of sensitives killed is directly proportional to the concentration of paramecin. This, confirming evidence of different types by AUSTIN, proves a single particle of paramecin can kill a sensitive paramecium and permits computation of the concentration of paramecin in undiluted killer cultures (order of magnitude, 350 particles per cc). Approximately 98 percent of the particles of paramecin are adsorbed by an excess of paramecia in 78 minutes. More than 90 percent of paramecin disintegrates in 4 hours at 40.5°, 8 hours at 32°, 27 hours at 21°, and 50+hours at 11°. Kappa is also destroyed at rates varying with temperature. At 32° and higher, its rate of destruction exceeds its rate of production; at 27° and lower, the reverse is true; but at about 10° destruction again seems to exceed production. Thus, between 27° and 32° and possibly also near 10° are temperatures at which the two rates are equal; their minimum values are given by the fission rates at these temperatures. Under conditions in which the concentration of kappa decreases, two methods may be used to find the number of kappa particles per original killer cell.

SPARROW, A. H., Harvard University, Cambridge, Mass.: *Changes in sensitivity of chromosomes to X-ray breakage during microsporogenesis.*—It has previously been established that chromosomes of Trillium undergo changes in radiosensitivity during meiosis. This investigation has been continued and a fairly complete sensitivity curve, including most stages from early meiotic prophase through pollen grain resting stage, has been constructed. A standard dosage of 50 r was given at all stages. Sensitivity is determined by counting the number of fragments per 100 PMC. Meiotic prophase stages up to and

including diplotene were scored at AI. All later stages were scored at microspore metaphase. The data show a gradual increase of sensitivity from early meiotic prophase up to metaphase I. The curve then drops down again to a low at the tetrad (or very early resting) stage and starts up again during the resting stage. The results are interesting in that (1) the range of sensitivities is very large (about 1 to 27), and (2) that high and low sensitivity stages respectively correspond to stages of high and low concentrations of desoxyribose nucleic acid in the chromosomes.

SPARROW, A. H. and MARIAN REESE HAMMOND, Harvard University, Cambridge, Mass.: *Desoxyribonucleic-acid-containing bodies in the cytoplasm of pollen mother cells at early meiotic prophase.*—Feulgen-positive bodies are present in the cytoplasm of pollen mother cells in several genera of plants (Lilium, Trillium, Allium, Paeonia and others). They apparently originate in the nucleus or at the nuclear membrane and then migrate out into the cytoplasm. They are sometimes attached to the nucleus by fine strands, which may occasionally pass through cell walls. There is great variation in position, size and shape of these bodies and in their effect upon the morphology of the nucleus. In extreme instances their total volume in a given cell is greater than that of the nucleolus. In such cases the nucleus may stain more lightly than usual, indicating a disturbance in nucleic acid supply. This may result from competition between nucleus and cytoplasmic bodies for the same materials. It is suggested that the cytoplasmic bodies may function as a modification of the more common type of heterochromatin-nucleolus mechanism controlling nucleic acid synthesis. They could thus be instrumental in building up the high concentrations of ribose nucleic acid present in the cytoplasm of pollen mother cells at early meiotic prophase stages.

STEBBINS, G. LEDYARD JR., University of California, Berkeley, Calif.: *Artificial synthesis of old and new polyploid species in Bromus.*—The annual species *Bromus arizonicus* has 42 pairs of medium sized chromosomes. When crossed with the perennial *B. carinatus* (n = 28) a sterile hybrid is produced which normally forms 7 medium sized trivalents, 14 medium sized bivalents, 14 medium sized univalents, and 7 large univalents, the latter derived from *B. carinatus*. This latter species has four distinct genomes, ABCL, while *B. arizonicus* has six, of which two are similar, ABCC'DE. Experimental evidence shows that the ABC group exists in three South American species, *B. catharticus*, *B. haenkeanus*, and *B. stamineus*, all with 21 pairs. An F1 hybrid between *B. catharticus* and *B. Trinii* (n = 21) had up to 7 pairs at meiosis, and the 42 paired allopolyploid derived from it resembles *B. arizonicus*. Since the differences between the *B. catharticus-Trinii* allopolyploid and *B. arizonicus* parallel those between *B. catharticus* and *B. haenkeanus*, *B. arizonicus* is interpreted as derived from *B. haenkeanus* × *Trinii*. Two new allopolyploid species, *B. carinatus-Trinii* and *B. maritimus-Trinii*, both with the genomic formula ABCC'DEL, are vigorous and highly fertile, and their establishment under

natural conditions is being attempted. The latter shows promise for revegetating certain livestock ranges of California.

STONE, WILSON S., The University of Texas, Austin, Texas: *A study of recombination hybrids in the virilis species group.*—A number of F_2 and F_3 hybrid segregants which had one or more foreign chromosomes introduced from another species were tested for viability, fertility and egg hatch. Some classes are viable and fertile, others have reduced fertility, while still others are sterile. The egg hatch from recombination females varied with the combination. Certain recombination males were sterile because of the failure of the last steps in the formation of mature sperm from the sperm bundles. Some genes can replace their alleles in these crosses without detrimental effects, others cannot.

STRONG, L. C., Yale University School of Medicine, New Haven, Conn.: *The induction of germinal mutations by chemical means (methylcholanthrene).*—Approximately 50 germinal mutations involving coat color have been obtained in mice following the subcutaneous injection of methylcholanthrene (both parents injected at 60 days of age for eighteen generations). This is a mutation rate of 1 in 500 mice (25,000 mice used in experiment either receiving methylcholanthrene or continued as untreated descent from experimental animals). The controls of similar and unrelated origin, never subjected to methylcholanthrene have shown a mutation rate of 1 to 26,250 mice (eight mutations in 210,000 mice over a period of 27 years). In addition to these clear-cut cases of germinal mutations, there have also been obtained significant alterations in susceptibility to many induced tumors and these alterations of susceptibility have shown linkage with well established genes such as brown and piebald, and sex. Thus it is reasonable to conclude that the effect of methylcholanthrene on the induction of tumors may be comparable to the effect of methylcholanthrene upon the germ plasm. Embryological disturbances such as dextrocardia, situs inversus and identical twins have also been obtained in the descendants of mice receiving methylcholanthrene over a number of generations. An increased prenatal mortality, alterations in sexual maturity, fertility and fecundity have also been observed.—In two series of germinal mutations (of independent origin) obtained in this experiment, there has been found a graded series of (a) mutant black (from brown) (b) pale black (c) graphite (d) dark gunmetal (e) gunmetal (f) darker bronze (g) dark bronze (h) bronze (i) dark sepia (j) sepia (k) the original well established brown (l) chestnut (m) light brown (n) lighter brown (o) tan and finally (p) buff. Each of these mutants breeds true and they probably form a series of alleles at the brown-black locus. Many somatic mosaics have been obtained between two of the above mutant types, such as between chestnut and brown or brown and buff, etc. It is obvious that an unstable genetic state exists in these two lines one of which was an outcross between the methylcholanthrene injected series and C57 blacks, and the other, from a single litter of methylcholanthrene treated mice (without an

outcross) in which three black mutants occurred in seven young. Since a majority of the germinal mutations have occurred in these two descents (out of a total of eleven descents continued) the mutation rate from these two unstable genetic states is considerably higher than 1 in 500 estimated for the entire methylcholanthrene injected descent.—A demonstration of new mutant types of coat color in mice is also available.

SWANSON, C. P., Johns Hopkins University, Baltimore, Md.: *A consideration of the structure of the prophase chromosomes in the pollen tubes of Tradescantia.*—Evidence, both observational and experimental, is presented which demonstrates that the prophase chromosomes in the pollen tubes of Tradescantia are quadripartite as early as two hours after germination of the pollen grain on an artificial medium. These observations are discussed in the light of current cytological theories regarding chromosome structure and behavior.

VALENCIA, J., Indiana University, Bloomington, Indiana: *The cytology of structural changes involving specific loci in Drosophila.* (Work supported by American Cancer Society grant recommended by Committee on Growth, National Research Council)—A series of mutants, produced by irradiation of spermatozoa in MULLER'S experiments (described in parallel paper), were subjected to salivary examination. All of them involved specific loci for already known visible mutations, as shown by their manifestation in compounds. In all cases thus far studied in which the visible effect had been found combined with a completely linked recessive lethal effect, but no marked disturbance of crossing-over, cytological evidence of a minute deletion (deficiency) was obtained, whereas deletions were not found in non-lethal cases. The validity of the genetic method used for determining the frequency of such deletions at these loci was thereby confirmed. Figures and preparations of some of these cases will be demonstrated, as well as of cases of gross rearrangement affecting the same loci. The deletions serve to establish more exactly the salivary map loci of the genes affected, and to indicate the range of length of small deletions in the given regions, while the gross rearrangements give evidence concerning the spatial and qualitative aspects of the position effect. The work also gave opportunity for a more exact study of the KOSSIKOV repeat (figures to be presented).

VALENCIA, R. MARIE, Indiana University, Bloomington, Indiana: *Is there transmission of the effect of heterochromatin on variegation in Drosophila?*—It is known that extra heterochromatin in the nucleus tends to normalize the peculiar variegated, or mosaic, position effect of certain genes which, through chromosome rearrangement, have been placed in proximity to heterochromatin. NUZHDIN has reported that this normalizing effect is carried over, maternally and paternally, from one generation to the next. The present experiments, carried on in collaboration with H. J. MULLER, were designed to recheck these results on other material. A member of the series of dominant variegated

alleles of brown eye, *bw*^{V4}, is being employed. This is a paracentric inversion, whereby the brown^{+} gene has been placed next to the heterochromatin of the right arm of the second chromosome. We find its variations, because of their conspicuous and measurable nature, easier to study than those of the characters yellow and achaete, when associated with the scute^{8} inversion, reported on by NUZHDIN. Isogenic stocks are being utilized, since it is known, in general, that mosaically expressed position effects influenced by heterochromatin are easily modified by the rest of the genotype as well. The extra heterochromatin in our work is supplied by a special Y (of the "*sc*.Y^{l}" type), containing yellow^{+} as a marker, so that its presense or absence can be determined with certainty. For greater objectivity and quantitative treatment, the variations are subjected to colorimetric measurement, following pigment extraction of the groups of flies by the method of EPHRUSSI. As the experiment is still in progress, the results will not be reported until the meeting.

VILLEE, CLAUDE A., Marine Biological Laboratory, Woods Hole, Mass., and Harvard Medical School, Boston, Mass.: *A quantitative study of phenocopy production with monochromatic ultraviolet irradiation.*—Drosophila larvae and pupae of known ages were irradiated with light from a mercury-argon resonance lamp with a nickel-cobalt filter which gives almost pure ultraviolet of wavelength 2537 Å. The intensity of the beam was measured with a Hanovia ultraviolet meter; by altering the distance from the light source to the target, intensities ranging from 105 to 440 microwatts per sq. cm. were obtained. Irradiation times varied from 10 seconds to 30 minutes. An increase in the sensitivity of larvae to irradiation occurs between 50 and 100 hours after hatching and a decrease occurs in the first 24 hours after pupation. The phenocopies most frequently produced were abnormalities in the abdomen; others found included dumpy wings, fused or singed bristles, fused eye facets, abnormal thorax, folded or curled wings, etc. Almost all of them can be explained as due to the killing of cells by the ultraviolet light; only two, a doubling of the sex combs of the male prothoracic leg, and a single shoulder excrescence, suggest the pervasive changes in morphogenesis which occur much more frequently as a result of X radiation.

VILLEE, CLAUDE A., Marine Biological Laboratory, Woods Hole, Mass., and Harvard Medical School, Boston, Mass.: *A spectrophotometric analysis of the eye colors of Habrobracon.*—The eye pigments obtained from wild type, white, ivory, canteloup, dark carrot and dark red Habrobracon were compared with those from *Drosophila melanogaster.* The Habrobracon heads were dissected off, the antennae removed, and the pigments extracted with the solvents used by EPHRUSSI and HEROLD, 30 percent ethanol acidified to pH2 and absolute methanol acidified with one percent HCl. The light absorption of the various solutions was measured with a Beckman spectrophotometer in 10mμ intervals from 300 to 625 mμ. The results indicate that none of the Habrobracon stocks have a red pigment similar to the one in Drosophila but that all have a brown

pigment similar to the brown pigment of Drosophila. The Habrobracon pigment becomes yellow when oxidized with peroxide and pink when reduced with bisulfite. The various colors in the several Habrobracon mutant stocks are due to differences in the quantity of the pigment or in the degree of oxidation or reduction of the pigment and not to the presence of different pigments.

WARMKE, H. E., and GERMAINE LE CLERC WARMKE, Institute of Tropical Agriculture, University of Puerto Rico, Mayaguez, Puerto Rico: ***The experimental control of leaf and root differentiation.***—The fleshy toproots of the genus Taraxacum and some other milky-juiced composites are known to possess a remarkable regenerative capacity. These can be cut into segments as small as 1 cm in length, and within 10–12 days rosettes of leaves will differentiate from callus tissue formed from the proximal surfaces. Callus, and sometimes roots, will develop from the cut surfaces of the lower, or distal, ends. These regenerative capacities have proved extraordinarily useful in an experimental study of the factors responsible for leaf and root differentiation. It has been found that the direction of differentiation can be altered by chemical treatment. If the proximal surface is treated with indolebutyric acid in concentrations of 100–400 p.p.m., roots will develop; if it is left untreated, leaves will develop. In like manner, either leaves or roots can be made to grow from the distal end. If this end is treated with indolebutyric acid, abundant roots will be formed; if it is treated with substances known to reduce auxin content, such as ethylene chlorohydrin, leaf primordia will differentiate instead. It thus appears that auxin level is vitally important in determining whether roots or leaves differentiate from a given mass of callus tissue. If this mechanism proves to be a general one, it may be an important beginning in the understanding of how the various complex organs of a plant are built from cells with the same chromosome complement and the same genetic constitution.

WEIR, J. A., Iowa State College, Ames, Iowa: ***A source of genes for evolutionary progress.***—The classical concept of progressive evolution of a species assumes either the occurrence of favorable mutations of genes which already possess vital catalytic functions or acquisition of new gene loci. The extreme improbability of a favorable mutation in an essential gene to produce a new vital function and the ultimate limit to increase in gene number imposed by the size of the nucleus render both views untenable. But this impasse can be met. Whereas an essential gene is inexorably confined by the vital nature of its primary function, a neutral gene would be subject to no such restriction. It could mutate toward a new function without detriment to the host. It is hypothesized that neutral genes will arise whenever the end products of the species' own essential genes are substituted preformed as the result of the gene action of other forms which constitute the nutritional environment of the species. A neutral gene is one which has lost its essentiality to the host and is therefore free to become incorporated in a totally different reaction chain. This hypothesis will account for the pyramidal parasitism found in nature. It

does not demand an alteration in gene number since the species merely supplements its own gene activity with the products of genes found in other species. In the process the species acquires neutral genes which are potential materials for evolutionary progress, but the number of loci remains unchanged. Man, at the vertex of parasitism, may have gained ascendancy by such a process.

WHITING, ANNA R., and BERNICE GOLDIS, University of Pennsylvania, Philadelphia, Pennsylvania: *Dose-action curves for X-ray-induced dominant lethal and visible mutations in Habrobracon eggs.*—Data are being collected for visible mutation rate of eggs X-rayed in prophase I with doses ranging from 6,400 r to 37,200 r. The dose-action curve suggests a direct relation to dose, thereby agreeing with work of previous investigators on this subject. Percentages of surviving eggs producing visibles range from 3.7 for 6,400 r to 15.6 for 37,200 r. This curve is compared with that of dominant lethality previously published by senior author for prophase I. The latter is complex and tends to become exponential with fractionation of dose which increases resistance to treatment.

WHITING, P. W., University of Pennsylvania, Philadelphia, Pennsylvania: *Linkage groups in Habrobracon.*—The combined sex, *X*, cantaloup, *c*, crescent, *cr* and red, *rd*, groups are *Sk* -12- *r* (-35- *"short"* -13- *gl* -30?- *x* -10- *fu* (-40- *"sharp"*) -24- *sb* -42- *bl* -30- *le* (-?- *sfa*) (-5- *My*) -12- *c* (-20?- *dII95* -15?- *SII*) -14- *l* -3- *n* -7- *ho* (-13- *cw*) -8- *vl* -15- *ro* (-17- *Yf*) -12- *bu* -37- *cr* (-15- *dr*) -40- *co/sl* -40- *ct* -34- *rd* (-11- *Cv*) -40- *gy* (-7- *ac/el*). The orange, *o*, group is *k* -28?- *dw* -5?- *m* -11- *o* (-19.5- *pupae naked lethal*). The white, *wh*, group is *semilethal* -?- *bk* -25- *wh/pl* (-22- *at*) -9- *st*. The small-wings, *sw*, group is *sv* (-24- *sw*) -23- *td* -27- *ma*. Two other pairs of gene differences show incomplete linkage, *wa* -22- *br* and *ta* -40- *un2*, and one shows complete linkage, *pk/ew3*. (Intervals indicating crossover units are given between partially linked genes. Complete linkage is indicated by /. Parentheses designate that direction left or right is undetermined.) Linkages here reported are from a *very* small percentage of tests made, all others giving independent segregation.

WITSCHI, EMIL, and JOHN J. MAHONEY, State University of Iowa and University of Illinois: *The genetics of the female prostate in rats.*—In rats, female prostate glands may be induced by prenatal injections of male sex hormone. The glands also develop spontaneously in some females. Surveys of wild rats and of several unselected strains of laboratory rats show various frequencies of occurrence. By inbreeding and selection, it was possible to develop two strains of rats differing characteristically in female prostate occurrence. This in itself indicates that the presence of a prostate gland in a normal female rat is under genetic control. In the *Prostate Strain* (*PS*) the frequency was increased from an original 29 percent to 95 percent. On the other hand, prostate incidence in the *Non-Prostate Strain* (*NPS*) females has been reduced to 4

percent. No visible differences exist between the male prostates of these two strains. Reciprocal cross breeding between the two groups gives the following results:

F1	PS ♀ × NPS ♂	61 per cent females with prostates
	NPS ♀ × PS ♂	42 percent females with prostates

All back crosses of F1 with NPS parent stock give approximately 30 percent occurrence in the offspring. Presence or absence of prostatic lobes in F1 mothers has no influence on the frequency of occurrence of the gland in their offspring. The following conclusions are presented. The factor controlling the development of female prostates is probably monogenic. Quantitative alleles determine various degrees of penetrance. It is not sex-linked. It is incompletely dominant. Some results suggest that homozygous mothers may increase the manifestation of the character in their offspring. The genetic aspects of the transfer of secondary sex characters from one sex to the other will be discussed.

WOOLLEY, GEORGE W., and M. M. DICKIE, Roscoe B. Jackson Memorial Laboratory, Bar Harbor, Maine: *Fuzzy mice.*—In 1945 our attention was called to a probable mutation to fuzzy in the CFW stock of F. G. CARNOCHAN at Carworth Farms. Some of the mutant mice were obtained from Carworth Farms and are being observed. Up to the present time the following breeding results have been obtained. In crosses fuzzy × fuzzy the offspring, 56 in number, were all fuzzy. In crosses fuzzy × normal all the offspring were normal, while in F1 × F1, 20.7 percent of 386 animals classified were fuzzy. In the backcross series, F1 × fuzzy, 50 percent of 236 animals classified were fuzzy. These data suggest fuzzy is a unit recessive gene. The symbol *fz* is suggested.

SPECIAL SUBSCRIPTION RATE FOR GRADUATE STUDENTS

A special rate of Three Dollars per current volume of GENETICS is offered to *bona fide* Graduate Students. A statement from an official of the institution certifying Graduate Student status should accompany subscription orders.

JOURNAL *of* GENETICS

Edited by

R. C. PUNNETT

and

J. B. S. HALDANE

Founded in 1910, the Journal of Genetics is the only British periodical devoted to the publication of original research in Heredity and Variation. Up to January, 1945, forty-six volumes have been completed. The illustrations form a feature of the series, for, in addition to numerous text figures and diagrams, the forty-six volumes contain 933 plates, of which 175 are in colors.

Many of the papers published deal with animals and plants of high economic value, and are therefore of interest to those concerned with agriculture and horticulture.

A full index of Vols. XXV-XXXVI was issued with the last part of Vol. XXXVI.

The Journal of Genetics is published in parts, of which three form a volume.

The Cambridge University Press has appointed The University of Chicago Press agent for the sale of the Journal of Genetics in the United States of America, and has authorized the following prices: Annual subscription $10.00 net; single copies $3.50 net each. The parts are sent post free to subscribers as issued.

Inquiry as to back numbers should be made to The Cambridge University Press. Separate parts and volumes are still available in some cases.

ECOLOGICAL MONOGRAPHS

(Issued under the auspices of the Ecological Society of America)

First issue published December, 1930

Managing Editors: A. S. PEARSE and C. F. KORSTIAN, *Duke University*

A quarterly journal devoted to the publication of original researches of ecological interest from the entire field of biological science. The journal will work in close cooperation with *Ecology* and will undertake the publication of papers of from 25 to 100 printed pages in length while *Ecology* will continue to specialize on papers of about 20 printed pages or less.

The board of editors will be glad to consider thoroughly scientific manuscripts which deal with any aspect of ecological investigation broadly interpreted and including community studies, ecological physiology, phenology, oceanography, biogeography, and ecological data from such practical fields as horticulture, agrioecology, economic entomology, forestry and fisheries, but will not include papers dealing primarily with economic problems.

Published in March, June, September, and December

SUBSCRIPTION: $6.00

THE DUKE UNIVERSITY PRESS

COLLEGE STATION

DURHAM, N.C.

Your Biological News

You would not go to the library to read the daily newspaper—probably you have it delivered at your home to be read at your leisure. Why, then, depend upon your library for your biological news?

Biological Abstracts is news nowadays. Abstracts of all the important biological literature are being published promptly—in many cases before the original articles are available in this country. Only by having your own copy of *Biological Abstracts* to read regularly can you be sure that you are missing none of the literature of particular interest to you. An abstract of one article alone, which you otherwise would not have seen, might far more than compensate you for the subscription price.

Biological Abstracts is published in eight low priced sections, as well as the complete edition, so that the biological literature may be available to all individual biologists. Section A, which includes abstracts of the genetics literature, is only $4 per volume. Ask for a sample copy.

BIOLOGICAL ABSTRACTS

University of Pennsylvania, Philadelphia 4, Pa.

PLANTS & GARDENS

New Non-technical Journal Published Quarterly by the Brooklyn Botanic Garden

Volume 1—1945
Volume 2—1946

Vol. 2. Spring Issue: Trees for Special Uses—Tree Culture.
Summer Issue: Roses—their culture—New Varieties.
Autumn Issue: Wild Flowers, a new approach.
Winter Issue: Digest of Year's Best Garden Articles.

1 year $2.00—2 years $3.00

BROOKLYN BOTANIC GARDEN
1000 Washington Ave., Brooklyn 25, N.Y.

NEW PUBLICATIONS RECEIVED

Scheinfeld, Amram. Women and Men. Pp. XX + 453. illus. Harcourt, Brace and Company, N.Y. October 1944. $3.50.

Small, James. pH and Plants, An introduction for beginners. Pp. VIII + 216. Figs. 16. D. Van Nostrand Company, N.Y. August 1946. $4.00.

Snyder, Laurence H. The Principles of Heredity, Third Edition. Pp. XVI + 450. Figs. 155. D. C. Heath and Company, Boston. February 1946. $3.75.

Wilson, Helen Van Pelt. Geraniums, Pelargoniums For Windows and Gardens. Pp. XVIII + 248. M. Barrows and Company, Inc., N.Y. June 1946. $2.75.

Westcott, Cynthia. The Gardener's Bug Book, 1000 Insect Pests and Their Control. Pp. XVI + 590. Figs. 105, 38 Color Plates. Doubleday & Co., N.Y. August 2, 1946. $4.95.

Weidenreich, Franz. Apes, Giants, and Man. Pp. VIII + 122. Figs. 90. University of Chicago Press, Chicago, Ill. July 15, 1946. $2.50.

Publications of The Wistar Institute

JOURNAL OF MORPHOLOGY

Devoted to the publication of original research on animal morphology and physiology, including cytology, protozoology, and the embryology of vertebrates and invertebrates.
Issued bimonthly, 2 vols. annually: $20.00 Domestic, $21.00 Foreign, per year.

THE JOURNAL OF COMPARATIVE NEUROLOGY

Publishes the result of original investigations on the comparative anatomy and physiology of the nervous system.
Issued bimonthly, 2 vols. annually: $15.00 Domestic, $16.00 Foreign, per year.

THE AMERICAN JOURNAL OF ANATOMY

Publishes the results of comprehensive investigations in vertebrate anatomy—descriptive, analytical, experimental.
Issued bimonthly, 2 vols. annually: $15.00 Domestic, $16.00 Foreign, per year.

THE ANATOMICAL RECORD

For the prompt publication of concise original articles on vertebrate anatomy, preliminary reports; technical notes; critical notes of interest to anatomists and short reviews of noteworthy publications.
Issued monthly, 3 vols. annually: $22.50 Domestic, $24.00 Foreign, per year.

THE JOURNAL OF EXPERIMENTAL ZOOLOGY

Publishes papers embodying the results of original researches of an experimental or analytical nature in the field of zoology.
Issued 9 times a year, 3 vols. annually: $22.50 Domestic, $24.00 Foreign, per year.

AMERICAN JOURNAL OF PHYSICAL ANTHROPOLOGY

Publishes original articles on comparative human morphology and physiology as well as on the history of this branch of science and the techniques used therein. In addition it gives comprehensive reviews of books and papers, an annual bibliography, and informal communications.
Issued quarterly, 1 vol. annually: $6.00 Domestic, $6.50 Foreign, per year.

JOURNAL OF CELLULAR AND COMPARATIVE PHYSIOLOGY

Publishes papers which embody the results of original research of a quantitative or analytical nature in general and comparative physiology, including both their physical and chemical aspects
Issued bimonthly, 2 vols. annually: $10.00 Domestic, $11.00 Foreign, per year.

THE JOURNAL OF NUTRITION

Publishes original researches in the field of nutrition and occasional reviews of literature on topics with which the journal is concerned.
Issued monthly, 2 vols. annually: $10.00 Domestic, $11.00 Foreign, per year.

For further information address

THE WISTAR INSTITUTE OF ANATOMY AND BIOLOGY
Woodland Avenue and Thirty-sixth Street **Philadelphia 4, Pa.**

Stain Technology

A Journal for Microtechnic

The latest developments in staining and general microscopy are presented in two different ways:

Original articles, 30-40 each year.

Abstracts of articles appearing elsewhere, so worded that the technic may be followed without consulting the original paper.

Issued quarterly **$3.50 a year**
20-year index **$1.50**

Just Out

5th Edition

Biological Stains

By H. J. Conn, Biological Stain Commission

Cloth, 5½ by 8¼ in.
346 pages

Price $4.25
(If cash companies order: $3.75)

For sale by

BIOTECH PUBLICATIONS
Lock Box 353, Geneva, N.Y.

ANNALS OF EUGENICS
A JOURNAL OF HUMAN GENETICS

Edited by L. S. PENROSE, assisted by
JULIA BELL, R. A. FISHER, J. B. HALDANE, MARY N. KARN,
R. R. RACE, J. A. FRASER ROBERTS

Contents of Vol. XIII, Part 3, November 1946

J. Grieve and G. M. Morant	Records of eye colour for British populations and a description of a new eye-colour scale.
H. Harris	The inhritance of premature baldness in men.
C. D. Lee and L. S. Penrose	A contribution to the genetics of hair colour in men.
D. J. Finney	Orthogonal partitions of the 6 x 6 latin squares.
J. B. S. Haldane	The interaction of nature and nurture.
B. S. Holt and M. E. Wright	A further note on polydactyly in mice.
D. S. Falconer	Sensory thresholds for solutions of phenyl-thio-carbamide.
R. A. Fisher	Note on the calculation of the frequencies of *Rhesus* allelomorphs.
J. E. H. Sawyer	A myopathic family.

Subscriptions, in advance, 50s. per volume.
Four parts, obtainable separately at 15s. each. Back volumes 55s. per volume.

Orders should be placed with

THE GALTON LABORATORY
UNIVERSITY COLLEGE, GOWER ST., LONDON, W.C.1.

Heredity and Variation in Microorganisms

COLD SPRING HARBOR SYMPOSIA ON QUANTITATIVE BIOLOGY

Volume XI (1946), about 300 quarto pages, 16 halftone plates, and 88 figures.

Problems of heredity in viruses, bacteria, fungi, and protozoa are considered in 27 papers written by: T. F. Anderson; D. Bonner; M. I. Bunting; M. Delbruck and W. T. Bailey, Jr.; M. Demerec and R. Latarjet; L. Dienes; R. J. Dubos; A. D. Hershey; A. Hollaender and C. W. Emmons; T. Johnson; J. G. Kidd; J. Lederberg and E. L. Tatum; C. C. Lindegren and G. Lindegren; S. E. Luria; A. Lwoff; E. C. MacDowell; M. McCarty, H. E. Taylor, and O. T. Avery; N. W. Pirie; G. Pontecorvo; M. M. Rhoades; O. W. Richards; F. J. Ryan; A. Shapiro; T. M. Sonneborn; S. Spiegelman; E. L. Tatum; and C. B. van Niel.

Price: $6.00 plus postage (domestic, 25¢; foreign, 50¢). Prepublication orders received before February 1, 1947 will be sent postpaid. Table of contents sent on request.

Address Biological Laboratory, Cold Spring Harbor, New York.

LA CELLULE

Recueil de Travaux originaux de Cytologie, Biologie et Histologie générale

Fondé par J. B. Carnoy en 1884

Publié par P. Debaisieux et P. Martens, professeurs à l'l'Université de Louvain (Belgique).

Trois fascicules constituent un tome généralement annuel, de 400 à 450 pages in quarto, avec nombreuses planches hors-texte en double page, noires et en couleurs, et des figures de texte.

Prix de souscription: Frs belges 450. port inclus.

Tables générales pour les tomes 1-40 (1885-1931); 100 pages in 4°; prix, séparément: Frs belges 100. Adresser les souscriptions à l'éditeur: Librairie Universitaire, rue de la Monnaie, Louvain.

Pour toute autre communication (rédaction, administration, échanges, publicité, etc.) s'adresser au Prof. P. Martens, rue Marie-Thérèse, 23 Louvain (Belgique).

THE EIGHTH INTERNATIONAL CONGRESS OF GENETICS will be held in Stockholm in the summer of 1948. The date will be announced later, when more is known about other congresses planned for 1948. Swedish geneticists have formed an Organization Committee for the Congress, with Professor Gunnar Dahlberg, of the University of Uppsala, as chairman, and Professor Gert Bonnier, Institutet för Husdjursförädling, Wiad, Eldtomta, as general secretary.

FAMILIAL SUSCEPTIBILITY TO TUBERCULOSIS

Its Importance As a Public Health Problem

By

RUTH RICE PUFFER, DR. P. H.

EXTRACTS FROM THE REVIEWS

René J. Dubos, in the *Harvard Medical Alumni Bulletin.*

"It [tuberculosis] constitutes the greatest single cause of death between the ages of 15 and 35. . . . There is much evidence, furthermore, that a recrudescence of tuberculosis will follow in the wake of the War. . . . It is gratifying, therefore, to find in Dr. Puffer's monograph a most valuable addition to our knowledge of the natural history of tuberculosis."

From *Child Development Abstracts and Bibliography.*

"The thesis . . . is that tuberculosis is a familial disease in the broad biological meaning of the term familial. . . . Aside from the careful statistical analysis which characterizes the work presented here, this book has the merit of bringing forcibly to the attention of public health authorities the importance of familial studies."

From the *Journal of Laboratory and Clinical Medicine.*

"Dr. Puffer presents her argument in a simple and convincing manner. Her book should prove as stimulating and helpful to the clinician as to the public health physician. It is a worthy companion to the other Harvard University monographs in medicine and public health."

AT ALL BOOKSTORES · SECOND PRINTING. $2.00

HARVARD UNIVERSITY PRESS

CAMBRIDGE 38, MASSACHUSETTS

INFORMATION FOR CONTRIBUTORS

Contributions to GENETICS may be in the field of genetics proper, of cytology, taxonomy, embryology, physiology, biometry, or mathematics, if of sufficient importance and of such a character as to be of primary interest to the geneticist. For the present, the length of manuscripts will be limited to twenty-five printed pages (about twelve thousand words) except by special vote of the Editorial Board. Tabular matter in excess of one-fourth of the manuscript cannot be printed, unless of particular importance, but will be kept on file for reference on request provided two copies are furnished by the author. Excess pages will be printed if paid for by the author (about six dollars a page).

Manuscripts are printed, ordinarily, in the order of their receipt. They may be printed out of turn provided the entire cost is paid by the author. Such material will be added to the current number and will not delay the publication of any other article.

Contributors are requested to use care in the preparation of manuscripts. Carbon copies cannot be considered. All references to literature should cite the name of the author, followed by the year of publication, the papers so referred to being collected into a list of "LITERATURE CITED" at the end of the article. In this list care should be taken to give the titles in full, and to indicate accurately, in Arabic numerals, the volume number, the first and last pages, and the date of publication of each paper if published in a periodical, and the number of pages, place and date of publication, and the name of publisher, of each independent publication. The arrangement of this list should be alphabetical by author and chronological under each author. Titles of publications are abbreviated according to the *World list of scientific periodicals*, Oxford University Press, London and New York, 1925.

Each manuscript should include a summary of the evidence and of the conclusions. Factor symbols should be separated so that they can be properly identified and underlined for italics. Gene symbols with sub- or superscript letters should be avoided except in long allelic series. Most typewriters do not distinguish between the letter l and the figure 1 or the hyphen and the dash. Such distinctions should be made wherever there is a possibility of confusion.

Footnotes should be avoided wherever possible. Usually, they can be enclosed in parentheses and inserted after the sentences to which they apply. If used in the text they should be numbered consecutively in a single series and designated by Arabic superscript numerals. Footnotes to tables should be marked with an asterisk, dagger, or other symbol so as not to be confused with the figures in the tables.

Illustrations should be referred to as figures wherever possible. Plates are reserved for illustrations that require paper inserts or for collections of small figures that cannot be designated conveniently as separate figures. Text figures should be clearly identified but not numbered in the illustration. Figures included in plates should be distinguished by letters rather than by numbers. All figures and plates are reduced to a maximum of $4\frac{3}{8}$ inches in width and 6 inches in height.

Legends for figures and plates should be typewritten separately from the illustrations for the reason that the type is set by the printer and the illustrations are made by the engraver. Mistakes are likely to occur if the separation is not made by the author.

Galley proofs and, whenever there is time, page proofs will be sent. Authors should leave forwarding directions whenever they are to be away from the address sent with the manuscript. Ordinarily page proofs cannot be sent out of the country. Both proofs must be returned promptly, and no extensive change may be made in page proofs which is not compensated for within the same paragraph or in an adjacent paragraph on the same page. Changes from copy will be charged to the author.

GENETICS furnishes 75 reprints, without covers, free. Covers and additional reprints may be secured at actual cost of manufacture provided these are ordered when the corrected galley proofs are returned.

Manuscripts and all editorial correspondence should be addressed to the Editor of GENETICS, 310 Dewey Hall, University of Rochester, Rochester, N. Y.

GENETICS Vol. 32 No. 1 JANUARY, 1947

CONTENTS OF PRECEDING ISSUES

MARCH, 1946

GARDNER, ELDON J., Sexual plants with high chromosome number from an individual plant selection in a natural population of guayule and mariola.

WRIGHT, SEWALL, and TH. DOBZHANSKY, Genetics of natural populations. XII. Experimental reproduction of some of the changes caused by natural selection in certain populations of *Drosophila pseudoobscura*.

SNELL, GEORGE D., An analysis of translocations in the mouse.

COOPER, KENNETH W., The mechanism of nonrandom segregation of sex chromosomes in male *Drosophila miranda*.

TAN, C. C., Mosaic dominance in the inheritance of color patterns in the lady-bird beetle, *Harmonia axyridis*.

Abstracts of papers presented at the delayed 1945 meetings of the Genetics Society of America.

MAY, 1946

WILSON, G. B., Cytological studies in the Musae. I. Meiosis in some triploid clones.

STURTEVANT, A. H., On the dot chromosomes of *Drosophila repleta* and *D. hydei*.

DOBZHANSKY, TH., Genetics of natural populations. XIII. Recombination and variability in populations of *Drosophila pseudoobscura*.

SHULL, A. FRANKLIN, Inheritance in lady beetles. IV. The form of the chitinous male genitalia in crosses of the species *Hippodamia quinquesignata* and *H. convergens*.

MOORE, JOHN A., Incipient intraspecific isolating mechanisms in *Rana pipiens*.

RUSSELL, ELIZABETH S., A quantitative histological study of the pigment found in the coat-color mutants of the house mouse. I. Variable attributes of the pigment granules.

JULY, 1946

KAUFMANN, BERWIND P., ALEXANDER HOLLAENDER, and HELEN GAY, Modification of the frequency of chromosomal rearrangements induced by X-rays in Drosophila. I. Use of near infrared radiation.

KAUFMANN, BERWIND P., and ALEXANDER HOLLAENDER, Modification of the frequency of chromosomal rearrangements induced by X-rays in Drosophila. II. Use of ultraviolet radiation.

STADLER, L. J., Spontaneous mutation at the *R* locus in maize. I. The aleurone-color and plant-color effects.

BLANC, RICHARD, Dominigenes of the vestigial series in *Drosophila melanogaster*.

GERSTEL, D. U., Inheritance in *Nicotiana Tabacum*. XXI. The mechanism of chromosome substitution.

VILLEE, CLAUDE A., Phenogenetic studies of the homoeotic mutants of *Drosophila melanogaster*. IV. Homoeotic and "growth rate" genes.

STRANDSKOV, HERLUF H., and EARL W. EDELEN, Monozygotic and dizygotic twin birth frequencies in the total, the "white" and the "colored" U. S. populations.

SEPTEMBER, 1946

KAUFMANN, BERWIND P., Modification of the frequency of chromosomal rearrangements induced by X-rays in Drosophila. III. Effect of supplementary treatment at the time of chromosome recombination.

CASPARI, ERNST, On the effects of the gene *a* on the chemical composition of *Ephestia kühniella* Zeller.

WILSON, G. B., Cytological studies in the Musae. II. Meiosis in some diploid clones.

WILSON, G. B., Cytological studies in the Musae. III. Meiosis in some seedling clones.

CHAPMAN, ARTHUR B., Genetic and nongenetic sources of variation in the weight response of the immature rat ovary to a gonadotrophic hormone.

NOVITSKI, E., Chromosome variation in *Drosophila athabasca*.

LOWE, JEANNETTE, and OLIVER E. NELSON, JR., Miniature seed—a study in the development of a defective caryopsis in maize.

NOVEMBER, 1946

DUBININ, N. P., and G. G. TINIAKOV, Inversion gradients and natural selection in ecological races of *Drosophila funebris*.

PAVAN, C., Chromosomal variation in *Drosophila nebulosa*.

TAN, C. C., Genetics of sexual isolation between *Drosophila pseudoobscura* and *Drosophila persimilis*.

MEAD, S. W., P. W. GREGORY, and W. M. REGAN, Deleterious recessive genes in dairy bulls selected at random.

MAMPELL, KLAUS, Genic and nongenic transmission of mutator activity.

STERN, CURT, R. H. MACKNIGHT, and M. KODANI, The phenotypes of homozygotes and hemizygotes of position alleles and of heterozygotes between alleles in normal and translocated positions.

HERSHEY, A. D., Mutation of bacteriophage with respect to type of plaque.

Summaries from forthcoming papers.

Index to Volume 31.

SINGLE NUMBERS $1.25 ANNUAL SUBSCRIPTION $6.00

obtainable from

BROOKLYN BOTANIC GARDEN, BROOKLYN, NEW YORK